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### **Review of the Potential Near-and Far-Field Effects of the Organic Extractive Component of Integrated Multi-Trophic Aquaculture (IMTA) in Southwest New Brunswick with Emphasis on the Blue Mussel (*Mytilus edulis*)**

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## Foreword

This series documents the scientific basis for the evaluation of aquatic resources and ecosystems in Canada. As such, it addresses the issues of the day in the time frames required and the documents it contains are not intended as definitive statements on the subjects addressed but rather as progress reports on ongoing investigations.

Research documents are produced in the official language in which they are provided to the Secretariat.

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## ABSTRACT

Research on and development of Integrated Multi-Trophic Aquaculture (IMTA) has been ongoing in the Bay of Fundy over the last decade. The industrial development of this concept has resulted in some questions related to the impacts and benefits of IMTA, such as:

1. What are the factors influencing the effects of IMTA?
2. Does IMTA help reduce the benthic organic loading at an aquaculture site?;
3. Does the current scale of IMTA in Southwest New Brunswick reap any benefits? If not, at what operational scale (i.e. spatial scale), species mix, and biophysical attributes could result in a net environmental benefit? (e.g. a reduction in the rate of carbon deposition); and
4. At what scale would IMTA have measurable impacts on other aspects of the ecosystem, such as on phytoplankton and on existing clam beaches?

There are several factors that determine the efficiency of open-water IMTA, including biological factors, such as species selection, the potential efficiency of the organism, species physiology, diet availability, and how efficiently the trophic linkages are established. The efficiency of those linkages strongly depends on the physical factors at the culture site, such as water depth, currents, and temperature. In addition, there will be a number of intentional and unintentional interactions within the ecosystem. These interactions can take the form of disease and parasitism at the small scale to predation events at the large scale. Understanding the scale and mechanisms of these ecological interactions will help to optimize outcomes and assist in the development of new management policies for IMTA. The more efficient the IMTA system, the less nutrients (energy) will be available for transfer to the outside environment.

Estimates from this study indicate that diets of extractive trophic level (ETL - blue mussels in this case) must consume 10 to 20% food from the fed trophic level (i.e. salmon culture solids) before there is a decrease in the net organic loading at an IMTA site. Whether an ETL can accommodate this amount of diet depends on the size classes of diet available and the ability of the ETL to exploit it. Several studies have shown that some open-water extractive species can utilize fish farm waste nutrients at a level of approximately 30% of their diet. To date, empirical field data on the blue mussel suggests these animals are capable of assimilating similar amounts of fish-derived nutrients in their diets as long as they are in contact with the farm derived food. The organic content of the waste from the ETL will be significantly lower compared to the original source, which makes a large difference to the loading of organic carbon to the bottom. Mussel faeces settle at a much slower rate and contain much less carbon than salmon faeces, depositing less carbon per unit area, and consequently resulting in a lower relative benthic impact. However, it is considered very unlikely that IMTA, using only fine particulate feeders (mussels) in Southwest New Brunswick, has significantly reduced loading of total particulate matter to the benthos at the sites.

Adopting IMTA can result in a number of costs and benefits. The present IMTA configurations are still in development, and, while their current configurations have likely not significantly lowered benthic loading at salmon farms, mussels have been used to remove fine particulates generated by salmon and kelp have extracted some fraction of soluble inorganic nutrients resulting from salmon metabolic and respiratory processes. Sea urchins and other species have the potential to be more effective at reducing benthic organic deposition beneath salmon farms. In the future, the design of the IMTA aquaculture sites will have to evolve both structurally and in complexity as the new sites will have to accommodate additional species and ensure that the flow of water is sufficient for the salmon, as well as efficiently connecting the various trophic levels. Calculations of the area available on existing aquaculture sites suggest that sufficient space is available on most existing leases to accommodate new IMTA modules. Projections on

potential production suggest that significant quantities of organisms could be grown and are theoretically capable of assimilating much of the nutrient fish waste (assuming appropriate contact rates). This level of production will require hatchery production of juveniles due to the volumes required.

There is the potential for IMTA operations to contribute to ecosystem stressors via nutrient waste addition and, in some scenarios, localized species addition. However, detecting small changes due to the IMTA beyond that of salmon aquaculture will be difficult. Some IMTA trophic levels, such as shellfish, that consume natural particulates in addition to fish farm wastes, may add increased nutrients under some circumstances; however, there is no evidence in Southwest New Brunswick that there is any nutrient limitation or eutrophication due to dissolved nutrients occurring except at very local transitory levels due to the high level of tidal flushing. It is not expected that there will be many changes to the phytoplankton on a broad scale, although there may be some local depletion levels around the farms due to filter feeders. The effects on secondary productivity through interactions with intertidal species are less certain. Depending on the hydrographic conditions of the intertidal zone, there could be interactions with the increased nutrients and epiphytic algae that create negative consequences for intertidal organisms. More research on larger ecosystem-scale effects is warranted.

**Examen des effets potentiels dans le milieu environnant et à distance de la composante d'extraction des éléments organiques de l'aquaculture multitrophique intégrée dans le sud-ouest du Nouveau-Brunswick, avec accent sur la moule bleue (*Mytilus edulis*)**

## RÉSUMÉ

La recherche et le développement de l'aquaculture multitrophique intégrée (AMTI) sont en cours dans la baie de Fundy depuis les dix dernières années. L'expansion industrielle de ce concept a soulevé certaines questions portant sur les répercussions et les avantages de l'aquaculture multitrophique intégrée, notamment :

1. Quels sont les facteurs qui influent sur les effets de l'aquaculture multitrophique intégrée?
2. L'aquaculture multitrophique intégrée aide-t-elle à réduire la charge organique benthique d'un site aquacole?
3. L'ampleur actuelle de l'aquaculture multitrophique intégrée dans le sud-ouest du Nouveau-Brunswick apporte-t-elle certains avantages? Si ce n'est pas le cas, quelle échelle d'exploitation (c.-à-d. échelle spatiale), quel mélange d'espèces et quels attributs biophysiques pourraient se traduire par un avantage net pour l'environnement (p. ex. une réduction du taux de dépôt de carbone)?
4. À quelle échelle l'aquaculture multitrophique intégrée aurait-elle des impacts mesurables sur d'autres aspects de l'écosystème, tels que sur le phytoplancton et sur les plages de myes existantes?

De nombreux facteurs déterminent l'efficacité de l'aquaculture multitrophique intégrée en eau libre, y compris les facteurs biologiques, comme la sélection des espèces, l'efficacité potentielle de l'organisme, la physiologie de l'espèce, la disponibilité de la nourriture et l'efficience avec laquelle les liens trophiques sont établis. L'efficacité de ces liens dépend grandement des facteurs physiques au site d'élevage, comme la profondeur de l'eau, les courants et la température. De plus, un certain nombre d'interactions intentionnelles et non intentionnelles au sein de l'écosystème entrent en ligne de compte. Ces interactions peuvent prendre la forme de maladies et de parasites à petite échelle ou d'événements de prédation à grande échelle. Une compréhension de l'échelle et des mécanismes de ces interactions écologiques permettra d'optimiser les résultats et contribuera à l'élaboration de nouvelles politiques de gestion pour l'aquaculture multitrophique intégrée en eau libre. Plus le système d'aquaculture multitrophique intégrée est efficace, moins les nutriments (énergie) seront disponibles aux fins de transfert dans l'environnement extérieur.

Les estimations découlant de cette étude indiquent que dans le cadre des régimes alimentaires de niveau trophique extractif (dans ce cas, il s'agit du niveau trophique extractif des moules bleues), l'alimentation doit aussi compter une consommation de 10 % à 20 % du niveau trophique nourri (c.-à-d. des déchets solides de la salmoniculture) avant que l'on dénote une diminution de la charge organique nette dans un site d'aquaculture multitrophique intégrée. La capacité d'un niveau trophique extractif à fournir un régime de cette envergure dépend de la taille de la nourriture disponible et de la capacité du niveau trophique extractif à en faire l'exploitation. De nombreuses études ont démontré que certaines espèces d'extraction en eau libre peuvent utiliser les nutriments émanant des déchets provenant d'exploitations aquacoles à un niveau d'environ 30 % de leur régime alimentaire. À ce jour, les données empiriques sur le terrain liées à la moule bleue laissent entendre que ces animaux sont capables d'assimiler des quantités semblables de nutriments émanant de poissons dans leur alimentation, tant qu'ils sont en contact avec les aliments d'origine agricole. Le contenu organique des déchets produits par le niveau trophique extractif sera beaucoup plus faible comparativement à celui de la source

originale, ce qui représente une grande différence pour la charge du carbone organique au fond. Les selles de moules se fixent beaucoup plus lentement et contiennent beaucoup moins de carbone que les selles de saumon. De plus, elles déposent moins de carbone par unité de surface et, par conséquent, leur incidence benthique relative est plus faible. Cependant, il est très improbable que l'aquaculture multitrophique intégrée, qui utilise seulement des organismes se nourrissant de particules fines (moules) dans le sud-ouest du Nouveau-Brunswick, ait permis de réduire de façon significative la charge de matières particulières totales des benthiques aux sites.

L'adoption de l'aquaculture multitrophique intégrée peut entraîner certains coûts et avantages. Les configurations actuelles de l'aquaculture multitrophique intégrée sont toujours en cours d'élaboration, et, même si les configurations actuelles n'ont pas vraiment diminué la charge benthique dans les exploitations salmonicoles, les moules ont été utilisées pour éliminer les particules fines générées par le saumon et le varech a permis d'extraire une fraction des nutriments inorganiques solubles découlant des processus métaboliques et respiratoires du saumon. Les oursins et d'autres espèces sont susceptibles d'être plus efficaces pour réduire le dépôt organique benthique sous les fermes d'élevage de saumons. À l'avenir, la conception des sites d'aquaculture multitrophique intégrée devra évoluer sur le point de vue de la structure et de la complexité, puisque les nouveaux sites devront s'adapter à d'autres espèces, garantir que le débit d'eau est suffisant pour le saumon et relier de façon efficace les différents niveaux trophiques. Les calculs de la zone disponible pour les sites aquacoles existants suggèrent que l'espace suffisant est disponible pour la plupart des baux en vigueur pour installer de nouveaux modules d'aquaculture multitrophique intégrée. Des prévisions sur la production potentielle laissent entendre que des quantités importantes d'organismes pourraient être implantées et que ces derniers sont capables d'assimiler, en théorie, une grande partie des déchets émanant de poissons (en supposant que les taux de contact sont appropriés). Ce niveau de production nécessitera la production d'écloserie de juvéniles, en raison de la quantité requise.

Il est possible que les opérations d'aquaculture multitrophique intégrée accroissent les agents de stress de l'écosystème en raison de l'ajout de nutriments émanant des déchets et, dans certains scénarios, de l'ajout localisé d'espèces. Cependant, la détection des petits changements découlant de l'aquaculture multitrophique intégrée au-delà de ceux de la salmoniculture sera difficile. Certains niveaux trophiques de l'aquaculture multitrophique intégrée (p. ex. mollusques) qui assimilent des particules naturelles en plus des déchets d'élevage pourraient se traduire par une hausse des taux de nutriments dans certaines circonstances. Toutefois, il n'existe aucune preuve dans le sud-ouest du Nouveau-Brunswick d'une limite ou d'une eutrophisation liée aux nutriments en raison de nutriments dissous, à l'exception des niveaux éphémères très locaux en raison du niveau élevé de renouvellement de l'eau par les marées. L'aquaculture multitrophique intégrée ne devrait pas entraîner beaucoup de changements sur les phytoplanctons à grande échelle, mais il pourrait y avoir un épuisement à l'échelle locale autour des sites aquacoles en raison des organismes filtreurs. Les effets de la productivité secondaire découlant d'interactions avec des espèces intertidales sont moins évidents. Selon les conditions hydrographiques de la zone intertidale, il pourrait y avoir des interactions entre les nutriments accrus et les algues épiphytiques, qui ont des conséquences négatives sur les organismes intertidaux. Une recherche plus poussée sur les effets sur les écosystèmes plus grands est justifiée.

## INTRODUCTION

Integrated Multi-Trophic Aquaculture (IMTA), previously known as polyculture, is an ecological approach to food production based within the field of ecological engineering. It is based on the principle of recapturing excess organic or inorganic nutrients that are produced from the feeding operations of one or more aquaculture species and subsequently partially recaptured through a series of other aquaculture organisms growing at different trophic levels. These trophic levels convert a particular nutrient from an organic to an inorganic form (or vice versa) while extracting some of the potential energy contained within via the cellular metabolic processes involved (Figure 1). Portions of the captured energy and nutrients, in the form of somatic tissue or skeleton, are removed from the aquaculture system through the harvest of the seaweed or animal with the intended result of decreasing the net nutrient loading to the ecosystem, increasing the profitability of the aquaculture operation to the farmer, and providing ecosystem managers with an active rather than a passive tool for managing the anthropogenic impacts to the environment. This approach is seen as more socially acceptable by the general public (Barrington et al. 2010) and fits into the overall strategy of increasing the environmental sustainability of the human food supply. The concept is applicable not only to open water net cage salmon culture, but can be used in offshore systems, closed containment systems, freshwater, and either intensive or extensive-based approaches. This concept has the potential to bridge both aquaculture and fisheries through the production of juveniles for reseeding initiatives, as well as the creation of additional habitat from the aquaculture infrastructure (Newkirk 1996).

Currently, on IMTA sites in the Bay of Fundy, the fed trophic level (FTL) is Atlantic salmon (*Salmo salar*) because it is the largest aquaculture biomass currently in Atlantic waters and, as such, readily enables the development of IMTA techniques. However, it should be recognized that the FTL could be any species, such as trout, charr, cod, halibut or shrimp, where additional energy, in the form of feed, is being added to the local environment. In some cases, the system could be designed around another extractive species that concentrates organic matter within a particular space. Filter feeders, for example, take small, low settling velocity particles from the water column, concentrate them into a food particle, which is then swallowed, some of the organic energy is extracted, and the remainder is egested and falls to the bottom. The faecal pellets from the filter feeders will still contain some organic matter that could be further utilized by other extractive species at the detritivore trophic level. Early IMTA experiments grew kelp in the effluent of blue mussels (Garabrand 1982).

Canada is currently one of the countries at the forefront of developing techniques for IMTA and has been actively studying and developing the concept since 2001 in both Atlantic Canada in the Bay of Fundy and on the west coast of Vancouver Island in British Columbia. The work has primarily been completed at the University of New Brunswick in Saint John and Fisheries and Oceans Canada (DFO) at the St. Andrews Biological Station and at the University of Victoria and the DFO Pacific Biological Station in Nanaimo. In 2009, the Canadian Integrated Multi-Trophic Network (CIMTAN), consisting of universities and other institutes, was established across Canada. This IMTA network created a large, multidisciplinary team tasked to advance the concept of IMTA. Today, the network is comprised of 26 scientists, 8 universities, 6 federal research facilities and 1 provincial independent research lab, covering 6 provinces and involving 3 industry research partners.

### Brief history of IMTA

The concept of aquaculture, in an ecological setting is not new. There are anecdotal reports that China was using freshwater ponds next to human agricultural activities to grow algae and fish

with the recycling of human and agriculture nutrients for human consumption. The first report on carp culture was written in 475 BC by Fan Li, and so the formalization of techniques for aquaculture goes back over millennia (Rabanal 1988). Anecdotal information exists for organisms being opportunistically co-cultured from ancient Egypt, France, and Italy. Brackish water pond culture in East Java was practiced as far back as the 1500s, when ponds known as "tambaks" were regularly closed off from the sea and the animals (juveniles of several species) trapped within them cultured and harvested (Brzeski and Newkirk 1997; Primavera 2000). Milk fish (*Chanos chanos*) were regularly produced in these ponds along with various species of crabs and shrimp using the semi-natural ecosystems.

In North America, the concept of ecological aquaculture or "marine polyculture" is generally acknowledged to have been initiated by Dr. John Ryther from the Woods Hole Oceanographic Institution (Ryther et al. 1972; Ryther et al. 1975; Ryther 1976; Ryther et al. 1977). The goal of his extensive team was to use the nutrient rich water being produced from secondary human sewage treatment plants in the Boston/Cape Cod area and extract the nitrogen using phytoplankton in a type of closed containment system. The phytoplankton would be fed to various filter-feeding shellfish species which would later be sold. Small juvenile fish and lobsters were released into the settlement ponds to consume the polychaetes and amphipods that fed on the solid wastes. Water leaving the facility was put through a macroalgal bed of seaweeds to extract the nitrogen produced from the various co-cultivated species to give the water a final "polishing". The results from their work demonstrated that algae could be grown quite easily and in high densities, particularly macroalgae as the microalgae were prone to sudden population crashes. The shellfish were also problematic as the phytoplankton strains that were initially being cultured were not suitable for the oysters (*Crassostrea virginica*) and hardshell clams (*Mercenaria mercenaria*) that were being used. Later improvements were made by changing the algal species, as well as, some of the shellfish species to Manila clams (*Tapes semidecussata*) and European oysters (*Ostrea edulis*), both exotic species.

The social and biological issues of eutrophication in marine systems in the late 1970s continued to drive research on how to create more sustainable food production systems in the marine environment (Tenore et al. 1974; Huguenin 1976; Tenore 1976; Hughes-Games 1977; Gordin et al. 1981). Much of the work continued on contained systems or closed-off natural ponds by members of Ryther's original team, and additional species within trophic levels were added, such as abalone, various polychaete worms, oysters and seaweeds such as *Ulva*. Other nodes of polyculture also began to develop in the late 1980s. A very prolific series of studies began in southern Israel focusing on contained, recirculating culture of sea bream (*Sparus aurata*) with biofilters of *Ulva* sp. and various invertebrates (Rabanal 1988; Krom et al. 1989; Krom and Neori 1989; Neori et al. 1989; Cohen and Neori 1991; Neori et al. 1991). At the same time, polyculture was being closely examined as a possible advanced aquaculture technique to increase food production in some parts of Asia and Africa (Bardach 1985; Bardach 1986) to solve food shortages.

In the early 1980s, an understanding of the life history requirements and cage culture technology improved to an extent that salmon aquaculture began to increase in various countries such as Norway, Scotland, and Canada (Liu et al. 2011). The large increase in fish production and levels of feed being consumed led to questions concerning the ecological sustainability of the salmon farming operations (Folke and Kautsky 1989; Kautsky and Folke 1989; Kautsky and Folke 1990; Folke et al. 1994; Kautsky et al. 1995; Newkirk 1996; Troell 1996; Chopin 1997). Concepts about the environmental footprint of salmon farming, how to deal with the nutrient load resulting from the feeding operations, and how the farming activity could fit in to the operation of the coastal zone all began to be developed around this time. A review on the subject (Brzeski and Newkirk 1997) led to the formulation of "sustainable coastal production

systems", a concept where both aquaculture and fisheries would be managed at the community level using ecological principles to partially manage the energy flows through the local ecosystem (Newkirk 1996).

In 2000, discussions were initiated in Canada between researchers and managers from the salmon aquaculture industry in both New Brunswick in the Bay of Fundy (Robinson-DFO and Chopin-University of New Brunswick) and British Columbia (Steve Cross-University of Victoria). In 2004, the term IMTA was coined to describe this type of ecological aquaculture to distinguish it from other forms of polyculture in the literature in which there may only be multiple species growing at the same trophic level and not creating any synergies between species. The term has since been adopted worldwide.

In 2001, experimental-scale research projects were initiated in the Bay of Fundy to test the applicability of growing additional species on commercial salmon sites. The success experienced in the early experiments resulted in scaling up the approach in 2003 to a small pilot-scale level on a single site. In 2007, larger industrial-scale work began at six farms that were involved in the production of the IMTA species of blue mussels (*Mytilus edulis*) and the seaweeds *Saccharina latissima* and *Alaria esculenta*.

## STATEMENT OF ISSUES

Recently, the development of the IMTA concept has reached a level where the public has expressed an interest in the IMTA products being produced (salmon, mussels and kelp), and the industry has developed enough technical expertise in IMTA practices to produce commercial quantities, although the largest market demand at the present time is still for the salmon. With the increase in demand, there was a corresponding increase in requests from the salmon farming industry to convert some of their traditional salmon aquaculture sites into IMTA sites. This increase in requests raised concerns from both provincial regulators and stakeholders in the coastal zone, such as the fishing industry, with regard to possible negative impacts on coastal benthic habitats and phytoplankton depletion. As a result of public meetings, a review of the IMTA concept, as well as some of the empirical data generated from the Bay of Fundy IMTA project was requested. The extractive species presently used in IMTA operations in SWNB include mussels and kelp, although preliminary studies have also investigated the use of sea cucumbers, sea urchins, and scallops. Much of the data used to address the questions posed at this meeting were based on results using the present IMTA configuration (mussels/salmon), which is still in pilot-scale development. The questions to be addressed by science were assembled by DFO Resource Management with input from other industry stakeholders.

### Objectives

The objectives of this Canadian Science Advisory Secretariat (CSAS) review were to evaluate the following questions:

- What are the factors influencing the effects of IMTA?
- Does IMTA help reduce the benthic organic loading at an aquaculture site?
- Does the present scale of IMTA in Southwest New Brunswick reap any ecological benefit? If not, at what operational scale (i.e. spatial scale), species mix, and with which biophysical attributes would be required to result in a net environmental benefit (e.g. a reduction in the rate of carbon deposition)?
- At what scale would IMTA have measurable impacts on other aspects of the ecosystem, such as on phytoplankton and on existing clam beaches?

## WHAT ARE THE FACTORS INFLUENCING THE EFFECTS OF IMTA?

There are several issues that need to be considered in evaluating the effects of IMTA. These issues can be grouped under the following categories:

- choosing the right species,
- efficiency of an organism,
- availability of diets,
- physical factors, and
- regulatory issues.

### Choosing the right species

The fed component of IMTA systems is referred to as the FTL (Figure 1) while the co-cultured extractive species comprise the extractive trophic level (ETL). Nutrient waste from the FTL can be partitioned into three categories and consequently, at least three different broad ETL groups or niches are required if all 'nutrient streams' are to be targeted. These nutrient categories and associated niches are as follows:

1. Dissolved inorganic nutrients from metabolic and respiration processes, or leached from solid organic waste, including forms such as ammonium ( $\text{NH}_4^+$ ) and orthophosphates ( $\text{PO}_4^{3-}$ ). Dissolved organic material (DOM), both colloidal and dissolved, is also produced by all trophic levels. Both categories can be absorbed by inorganic extractive species, such as seaweeds and aquatic plants, as well as, a portion of the microbes.
2. Small suspended or slow sinking organic particulates, generated from feed waste or faeces, behave similarly to neutrally-buoyant particles and can be 'captured' by organic extractive suspension-feeders, such as shellfish and some grazers.
3. Larger, settleable and highly organic solids such as large settling particles or aggregates are also generated from feed waste or faeces and can be consumed by organic extractive deposit-feeders, such as sea urchins, sea cucumbers, sea worms and bottom fish (e.g. mullets, flatfish).

The role of an IMTA species is to act as a component in an ecological engineering approach to efficiently convert available aquaculture nutrients from one form to another (organic to inorganic or vice versa). During this conversion, the potential energy is extracted and converted into body tissue where it is stored for later use by the organism or is released in the form of metabolic waste, where it is potentially available for use by other organisms.

An IMTA organism should possess the following attributes:

- Occupy a trophic niche that targets one of the available nutrient categories. Multiple size classes of organic nutrients are available and no one organism can effectively consume all size classes. Most organic extractive species are specialists that only eat a particular size range.
- Able to efficiently capture and convert consumed or absorbed nutrients into body tissue or metabolic waste.
- Able to be produced at volumes large enough to sequester significant quantities of available aquaculture nutrients. The organisms should be easy to produce in large quantities as juveniles and grown commercially within the IMTA engineered system without significant logistical problems.

- Have an inherent value to the farmer that contributes to the income stream of the business. This could either be a direct product sale, such as food or other biological products, or a service function that either increases the profitability of the IMTA products or reduces cost from another aspect of the operation (possibly increasing fish health, environmental monitoring requirements, etc.). If the species is being considered as a human food item, it would be beneficial to have an existing market demand in place with an appropriate marketing strategy prior to it being produced.
- Should be socially acceptable and not create societal or legal issues.

### Effectiveness of an organism

Organisms, in general, are poor converters of consumed energy. Most organisms retain less than 25% of the food they consume (Smith 1992). Fish and other poikilotherms ("cold blooded" organisms) are more efficient than most terrestrial animals because they are not required to maintain a constant body temperature (which is energetically expensive) and they expend less energy to compensate for the force of gravity. Because all IMTA organisms to date are poikilothermic, fish and other marine organisms make good candidates.

### Trophic Transfer Efficiencies and Open-Water IMTA: Application and Considerations

One of the major challenges of assessing the effectiveness of open-water IMTA systems is determining the proportion of nutrients removed by the organic extractive niches. Determining the fate of organics from the FTL through the organic extractive niche requires details on what has been delivered, ingested, digested, egested, excreted, and retained by co-cultured species. In the case of the inorganic extractive niche, where soluble nutrients are removed, it may be acceptable to remove nutrient equivalents of what has been loaded in the water through the farming operation. For example, removing a unit of ammonium from the water column will have the same result on the localized concentration regardless of whether it was excreted from fish or was part of the ambient concentration. Consequently, the amount of nutrient in harvested biomass can be juxtaposed with what has been loaded by the farm. However this approach is not easily applied to the organic extractive niche, since it is the specific solid load from a farm that needs to be targeted. This waste stream is more likely to have discrete near-field impacts such as those potentially associated with organic deposition beneath cages. Environmental responses to benthic loading such, as changes in hydrogen sulphide levels, are used for regulatory compliance in several jurisdictions (Hargrave et al. 2005; Chang et al. 2007).

After nutrient delivery, knowledge of the amount of organic material originating from the farm and ingested by organic extractive species is typically a prerequisite for determining the amount digested, egested, excreted, and retained (Figure 2). While it is possible to "back calculate" the amount of diet consumed with the proper data, it is arguably impractical in an open-water, leaky system. If, for example, the digestibility of the diet is known (e.g. absorption efficiency of organic material) and all the faeces are collected, the amount of diet consumed can be determined. Biomass of co-cultured species harvested in conjunction with tissue composition can indicate how much of a farm-based nutrient was retained, but only if farm-based organics were the only food source; a scenario unlikely with shellfish and other invertebrates. In nature, nutrient losses from diet acquisition by one species can be readily exploited by several others. Remoras feed on shark leftovers and some fish in tropical reefs eat faeces from upper trophic level fish (Bailey and Robertson 1982; Robertson 1982). IMTA copies nature in this respect. It is also important to note the limitations of tracers in this respect. Tracers such as stable isotopes or fatty acids can identify what proportions of different dietary sources were consumed, digested, and retained as an end point (assuming all sources are known), but used in isolation, cannot quantify the amount consumed.

Therefore, if it is impractical to determine key parameters such as amount of material consumed, other approaches need to be considered. Similar difficulties have been encountered when attempting to track nutrient and energy transfer through ecosystems. Trophic transfer efficiency (also known as ecological efficiency) is one metric that may have potential in IMTA systems. Trophic transfer efficiency is the amount of food available at one trophic level, divided by the biomass produced at the next (Smith 1992) using mass or energy units. Such an approach may have merit in open-water IMTA systems as a means to avoid intermediate measures between loading from the FTL and biomass response of co-cultured species. The classical order of trophic levels may not be applicable in IMTA systems, however, since the mechanisms of delivery and type of food may differ. Nevertheless, the equation of trophic transfer efficiency for a given nutrient becomes:

Equation 1:

$$\text{Efficiency}_{\text{Transfer}} = \frac{\text{Biomass}}{\text{Food}_{\text{Available}}} \times 100$$

Approaches determining the portion of biomass growth attributable to the FTL (e.g. cultured salmon) will depend on the co-culture species niche. If captive deposit feeders are held under cages in the plume of organic deposition, it can be assumed that most, if not all, of the changes in biomass, can be attributable to farm organics. In the case of shellfish, which will have access to their natural diet from the plankton, as well as particulates from the FTL, it will be the augmented growth attributed to the farm that is needed to calculate the efficiency. This augmented growth can potentially be estimated by comparing growth performance between an IMTA location and a reference site, assuming there are no confounding factors. This can be very difficult in areas with high densities of fish farms and potentially different hydrodynamic exposures on the scale of dozens of meters. In addition, the expense of setting up an independent reference site is often beyond the financial means of researchers.

Trophic transfer efficiency is one of the classical measures in ecology. The retention of a small percentage of the diet in body tissue (growth) indicates some material is uneaten, undigested, used as fuel for respiration and reproduction, and, therefore, not converted to biomass. Does this mean that open-water IMTA is subject to the same constraints? While the value of 10-20% is often used as a 'rule of thumb', mean transfer efficiencies can range from less than 5% to more than 30% depending on the trophic level and whether the medium is mass or energy (Odum 1983) (Table 1). It is important to note that the aforementioned values refer to a single trophic transfer. Multiple successive transfers of organic material across trophic levels will result in increased net biomass sequestration of the original source for the IMTA system. In a multi-niche IMTA system, there can be the potential for multiple transfers and, as a result, the amount of nutrient sequestered from the system will be additive. However, progressively less material from the original source is removed with each successive trophic transfer and in this respect, the fate of fish farm organics in ecosystems is no different (Gyllenhammar and Håkanson 2005). It is also important to consider that there are multiple pathways of trophic transfers and this is reflective of the different niches in an IMTA system. Trophic transfer efficiencies are associated with each of three primary IMTA waste streams; dissolved inorganic and organic (DOM), settleable organic, and particulate organic nutrients. Consequently, as more niches are filled, there are more opportunities for nutrient recapture and an increase in the system transfer efficiency.

Aquaculture provides an opportunity to achieve high levels of trophic transfer efficiencies by facilitating access to waste streams and selecting co-cultured species that readily digest or uptake the nutrient waste available. For example, using present day values from the salmon industry, if 100 kg of feed is offered to Atlantic salmon, with a feed conversion ratio of 1.1, and

3 kg of feed is wasted, there is a growth of 88.2 kg from the consumption of 97 kg feed. With moisture in modern feed and large Atlantic salmon of about 8.5% (Reid et al. 2009) and 63% (Bjerkeng et al. 1997) respectively, a dry biomass of 32.6 kg is produced from 91.5 kg dry diet offered. This results in a trophic transfer efficiency of approximately 36%, arguably higher than that typically found in nature. This value, however, is a function of decades of development on highly digestible feeds, genetic selection of efficient growing salmon and well-designed feeding regimens to minimize feed loss.

Feed for the FTL (e.g. cultured salmon) is typically formulated to optimize growth and health. Consequently, there is little opportunity to influence the composition of the resultant faeces as a food source. Feeding salmon is also a function of the logistics required to supply the biomass present and demand of the fish. Faecal generation will be a function of the type of fish feed consumed (Reid et al. 2009). Consequently, co-cultured species will not be fed in the same manner. If the aim of a farm is to reduce potential environmental impact, the biomass of co-cultured species must be appropriate to accommodate the waste nutrient load available. Also, waste nutrients can only be indirectly routed to co-cultured species by placing them at a location and distance that will be in the predominant waste stream. This is unlike fish feeding where feed is timed and directed to maximize consumption, and waste feed is minimized by stopping when the fish are sated (i.e. the usual practice). Trophic transfer efficiencies in the organic extractive niches, therefore, are likely to be much less than that of cultured fish. Nevertheless, trophic transfer efficiencies may provide a theoretical upper limit for expectations for single transfers. Given the "best case scenario" from fish culture and maximum transfer efficiencies found in nature, it is unlikely that single trophic transfers in open-water IMTA systems will exceed 30%.

While trophic transfer efficiencies have some merit being applied to open-water IMTA systems, caution is warranted when relying on this measure to infer sustainability. Trophic transfer efficiency only addresses material transferred to biomass, as opposed to other nutrient transformations or losses of soluble organic matter (DOM). Consequently, inorganic soluble nutrients are not included in classical measures of trophic transfer efficiency, thereby excluding the role of inorganic extractive species such as kelps. Some of the organic load consumed by IMTA co-cultured species will be metabolized and excreted. Digested proteins consumed in fish faeces will partially be catabolized and excreted as ammonium, and portions of the organic carbon will be respired as carbon dioxide (Reid 2007). This raises the additional question of whether the transformation of a nutrient reduces the potential for negative environmental impact and brings in issues of differences between near-field and far-field effects.

On Canada's East and West coasts, benthic hydrogen sulfides measured under or along transects from fish cages are used for regulatory compliance. From this regulatory perspective, only the reduction of excess organic carbon deposition is important. Is there an environmental benefit from the conversion of organic carbon to inorganic carbon (carbon dioxide)? Inorganic carbon, such as carbon dioxide, in water has not traditionally been considered an environmental concern. However, recent developments with ocean acidification due to increased atmospheric carbon dioxide levels (Denman et al. 2011) have warranted serious consideration. The carbon in carbon dioxide from respiring cultured fish will originate from the feed and varying portions of this will be of terrestrial origin. If one of the objectives of IMTA is to remove the inputs associated with aquaculture, soluble carbon may warrant further inclusion (including both CO<sub>2</sub> and DOC). This is a complex issue, but nutrient transformation may also need to be considered in addition to sequestration in harvestable biomass. In any case, a 'properly stocked' IMTA farm may have another niche capable of intercepting these transformed nutrients (such as the absorption of carbon dioxide by kelps).

IMTA shellfish growth can be augmented without the direct consumption of farm-based organic solids. A link may also exist between the dissolved inorganic nutrients excreted from fish and enhanced planktonic primary production; indirectly influencing shellfish growth (Sarà et al. 2012). This feedback effect may be more likely to occur in warmer climates in oligotrophic conditions and areas of slow currents and is less likely to occur in Canadian waters. In the Passamaquoddy Bay area, chlorophyll concentrations near fish farming locations are no different than elsewhere in the Bay (Martin et al. 2006; Lander et al. 2013). Nevertheless, the potential for multiple-routes to augment shellfish growth at fish cages does illustrate some of the complexities and diversity of mechanisms involved in the IMTA shellfish component.

### Available Diets

Consumption of organic solids from the FTL (e.g. salmon) will remove a portion of organic material from the total mass available and the subsequent egestion by the ETL will result in "secondary faecal" production with a lower amount of net organic material than the original dietary material. This can be considered a form of "organic stripping" within the system. However, in order to quantify the amount of organic stripping occurring, the proportion of diet originating from the FTL must be known since some ETLs, such as shellfish, will also filter natural diets. Once natural diets are consumed, the indigestible portion is incorporated into a faecal pellet, which then will have a much higher settling velocity in the water column compared to the original dietary particle. Therefore, some of the planktonic natural seston that would otherwise pass through a site without adding to the organic load from the farm now has the potential for redirection to the benthos as settleable organic material as faeces. This raises the question of, when does shellfish culture in IMTA systems result in a net removal or addition of organic material to the system and how efficient can these systems be?

Efficiencies in IMTA systems are, in part, a function of proportions or ratios of ETL production compared to production at the FTL (e.g. Atlantic salmon). Specifically, the proportion of the overall FTL load that is consumed by organic extractive species (or absorbed by inorganic extractive species in the case of seaweeds) needs to be determined. Previous work compared a number of diet and corresponding faecal organic contents and absorption efficiency for mussels on natural and salmon culture diets (Reid et al. 2010) (modified in Table 2; Table 3).

Current commercial salmon feed is formulated to be highly digestible ("nutrient dense"), producing minimal faecal production while producing maximum growth. Faecal production can be estimated with digestibility knowledge of the different nutritional categories. For every unit of feed consumed, approximately 15% will be egested as dry faeces. The proximate composition of salmon feed, the estimated composition and amount of faeces produced per unit of feed consumed is shown in Table 2, from Reid (2007).

As feed is shipped, small particulates called 'fines' are created through abrasion among feed pellets and container surfaces which settle in feed bags or totes. Fines are too small to be consumed by the fish and essentially become waste food. In the early stages of industry development, losses due to fines generation could be significant. Modern pellet extrusion techniques have reduced this substantially and it is recommended practice to use guaranteed feeds with maximum acceptable level of fines (FAO 2001; Tacon and Forster 2003).

Nevertheless, excessive 'handling' can still generate fines (Miller and Semmens 2002) and, consequently, a lot of effort has gone into feed delivery mechanisms to minimize the generation of fines. Fines are typically included under the classification of 'waste feed'. While the mass of fines is likely small relative to overall feed inputs and feed wastage, there is little published literature that addresses the amount of fines entering the water. However, this is the part of the salmon feed component that is potentially available to a portion of the ETL filter feeding species, and therefore warrants consideration.

In addition to the rate functions associated with the capture and processing of food, contact time of the ETL with food particles is another important factor affecting organism efficiency. Contact time is a function of both time and space. Nutrients generated from a particular trophic level on an IMTA site will be subject to advection away from that site by currents created by wind and tides and depending on their distribution and the degree of patchiness, there will likely be variable contact rates with the ETL. For example, ammonium, produced from anabolic processes of organisms consuming food on site, will be advected away from the source at a rate determined by the current speed. Assuming the nets of the cage system are reasonably clean, ammonia from salmon sites could be released from 1 to 15 m in the water column. Similar issues exist for the feed or faecal particles that are produced. A model was developed (Cranford et al. 2013) for mussel feeding and digestion physiology and used to predict the effectiveness of particulate fish waste capture and absorption by mussels under present and alternative IMTA site scenarios. The capture efficiency of fish waste particles by mussels is limited by the time available to filter the water as it flows past the IMTA mussel structures. This conclusion is also supported by the literature (Troell and Norberg 1998). Present configuration of IMTA systems employing mussels on a few rafts are predicted to be highly inefficient at intercepting wastes from fish net-pens. Model scenarios with intensive (high stocking densities) and spatially extensive mussel culture conditions indicate that it is highly unlikely that mussels are capable of capturing the majority of fish waste at salmon farms in SWNB.

### Physical factors

The physical features of the IMTA site play a major role in the flow of nutrients through the system and the options available to the farmer on the type of structures and operations that can be logically accommodated. Therefore, choosing the proper site characteristics is one of the first steps in designing a successful IMTA operation.

#### Depth

The depth of the water column in which the IMTA site operates determines a number of key parameters. Much of the focus of the IMTA concept is optimizing the contact rates of nutrients being transferred between different trophic levels. Water depth combined with the settling velocity of organic particles determines the amount of time a particle will remain in suspension and, in combination with the average current velocities, the degree of dispersion away from the source of origin of the particular nutrient. As water depth increases, particles are likely to be advected further before settling to the bottom. This results in less relative loading per unit area of organic carbon to the benthos since the fixed load of organic nutrients is being distributed over a larger area (e.g. g C/m<sup>2</sup>/day). Water depth is negatively correlated to the probability of re-suspension. Water column depth also plays a role in site logistics. Shallow sites (less than 30 m) can be accessed by SCUBA divers, which allow subsurface structures in the water column or on the bottom of the lease area to be created, monitored, maintained, and more easily harvested. While the use of divers is expensive, having the ability to observe a structure and the ability to make spot repairs is valuable. IMTA sites that are situated in deep water (greater than 30 m), such as those found in fjords or further offshore are still feasible; however, accommodation has to be made for engineering solutions that allow for the remote handling of equipment. If benthic structures are used, remote monitoring solutions such as drop cameras or remote operated vehicles (ROV's) can be implemented; however, they are not as efficient as a diver for the overall visualization of a structure and detection of potential problems. A benefit of deep-water sites is the provision of additional space for mid-water applications since fish cage depth is generally standard, resulting in more distance between the bottom of the net and the sea floor.

## Currents

Water current patterns are one of the key factors to consider in the selection of a site (Cranford et al. 2013). Stronger mean currents result in higher flux rates of material through the site; thus, potential food produced in the ecosystem outside the farm will be delivered at a higher rate to IMTA organisms that are capable of using it. Excess nutrients from the IMTA farm will also have a higher flux rate and, therefore, the dispersal of nutrients will increase in distance away from the point source. High current speeds are not necessarily advantageous when connecting trophic levels within the IMTA system. High current speeds minimize contact time of nutrients with the various IMTA trophic levels since trophic levels need to be contained within the spatial boundaries of the lease. This point has been raised by an IMTA model developed from some Swedish work on salmon and mussel culture (Troell and Norberg 1998). Therefore, higher currents are not necessarily better when considering the efficiency of an IMTA system since longer contact rates between trophic levels results in higher transfer efficiencies. This conclusion is contrary to the current strategy of finfish aquaculture. In salmon culture, for example, large sites benefit from placement in accessible areas with high current exchange, which reduces the accumulation of organics on the sea floor, ensures adequate oxygen supply, and potentially reduces the attachment potential for some external parasites such as sea lice. A compromise will have to be achieved to optimize the operation of IMTA sites.

In higher currents sites, flux rates may be mediated through engineered solutions where water movement is slowed through the installation or configuration of farm structures. For example, water flow has been shown to be significantly reduced in the middle of aquaculture farms for blue mussels in Norway (Strohmeier et al. 2005). Placing the extractive species in the middle of the farm configuration is one method of increasing the nutrient contact time between trophic levels. However, the mussel rafts also deflect a portion of the water flow around them due to the density of organisms within the structure, resulting in the potential loss of food to filter feeders.

Water currents also present logistic challenges to working in aquaculture sites. The forces associated with high volumes of water moving through the farm site are very large, and gear that is engineered for the site has to be able to meet minimal specifications to be maintained. In many cases, work around traditional salmon farms is scheduled around the tides so that the forces associated with moving equipment and replacing nets are minimized. Tidal currents would also have to be taken into account, particularly in the Bay of Fundy, when an IMTA component is integrated on the site.

## Temperature

Temperature will play a role in the degree of success of the IMTA operation. Since all the animals being considered are poikilothermic, increases in temperature, within certain ranges, will result in higher physiological rates such as feeding, digestion, excretion, and respiration. Optimal ranges for these functions will vary by species, but are anticipated to closely match the characteristics of the fed component (e.g. salmon). However, conversion and feeding rates will vary by season in temperate climates. Temperatures in the Bay of Fundy range from approximately 0° to 16°C; thus, significant changes in various physiological rates are expected. Temperature will also affect the ability to work on site. Lower temperatures tend to make materials brittle and in the winter, ice from freezing sea spray will periodically cover the structures and create large forces on the structural materials. Structures and the organisms contained within them need to be able to withstand these forces, as well as the currents created by the wind and tides.

## Oxygen

In the Bay of Fundy, the oxygen concentrations range seasonally from 7-13 mg/L (Page et al. 2005) and can occasionally be supersaturated in the spring due to phytoplankton blooms and colder water and under-saturated in the fall. Seasonal patterns can be modified in areas subject to eutrophication where the biological oxygen demand from the microbial community is increased due to large flux rates of organic carbon to the bottom subsequently lowering oxygen concentrations towards the sea bottom.

Replenishment of oxygen to an area is accomplished through the tidal transport of new water, as well as oxygen exchange through the air-water boundary. At peak flood or ebb tide, more water and hence oxygen will be advected through the site. Therefore, any significant oxygen depletion due to fish would likely be observed at slack tide when water exchange is at a minimum. While some studies support decreasing oxygen at slack tide, the phenomenon is site-specific and not seen at all sites (Wildish et al 1993; Page et al. 2005)

There may also be certain areas where oxygen demand is high due to high biomass, eutrophication, and associated microbial action. For example, in Black's Harbour, the site of a local fish processing plant in an enclosed harbor, oxygen concentrations can decrease to almost zero during the summer and fall (Wildish and Zitko 1991). In these cases, oxygen can be driven to much lower levels in the water, but these are generally the exceptions. For salmon farming and IMTA, oxygen is generally not considered an issue. Oxygen demand from salmon cages or a raft of mussels is small considering the total oxygen available, assuming a small flushing rate (Table 4). Site selection should avoid areas with low flushing rates and the potential for the accumulation of high levels of organic material resulting in high levels of biological oxygen demand (BOD). This could result in temporarily lowering oxygen concentrations in water, subsequently impacting IMTA organisms.

## Species Interaction

The interaction between the local, naturally occurring species in the vicinity of IMTA operations can be significant. Phytoplankton produced in the region represents a potential food resource for filter feeding organisms stocked on IMTA sites. If filter feeding organisms grew on the external surfaces of IMTA structures containing organisms that prey on them, natural phytoplankton could move through two trophic levels to be incorporated ultimately into tissues of IMTA species. Accounting for the effects of external food sources is complicated and generally requires the use of tracers such as stable isotopes or fatty acids (Fernandez-Jover et al. 2009; Henderson et al. 1997; Olsen et al. 2012; Redmond et al. 2010) or more integrated estimates, such as differential growth patterns in relation to reference sites. The effect of external primary food sources on IMTA operations can be mixed. From a production perspective, providing that the IMTA species can assimilate more food, additional food resources from natural sources can increase the growth rates of filter feeding species resulting in more biomass. From the nutrient recycling perspective, outside food resources effectively compete with the uptake of aquaculture nutrients resulting in lower conversion efficiencies of the nutrient load coming from the FTL.

Another interaction from external primary production would be the effects of harmful algal blooms (HABs). HABs have been shown to negatively affect salmon farming operations (Sephton et al. 2007) by killing fish. At certain concentrations, HABs have also been shown to reduce filtration rates of some filter feeders such as clams, implying that the conversion rates of IMTA nutrients will be reduced as well (Bricelj and Shumway 1998; Navarro and Contreras 2010). From a marketing perspective, the presence of paralytic shellfish poisoning toxin (PSP) prohibits any marketing of the product. Currently, the establishment of a closed marketing window during the PSP season avoids the marketing issue.

The secondary production of the natural ecosystem in proximity to IMTA sites also creates several interactions at multiple scales. At the microscale, the interactions of viruses and bacteria with the macrofaunal species play important economic roles although the processes and mechanisms involved are not particularly well understood. Pathogenic viruses and bacteria create disease situations within the IMTA organisms when the correct combination of host, pathogenic agent and environment are present. Diseases such as infectious salmon anemia (ISA) and bacterial kidney disease (BKD) have resulted in large economic losses to the Atlantic salmon industry. One concern from the fish health sector (e.g. veterinarians) is whether some IMTA species act as reservoirs for pathogenic agents and either initiate an infection cycle or prolong an existing one (Meyers 1984). The relevance of this concern is related to scale. If the probability of infection is proportional to the number of IMTA species being cultured, then the risk is increased as the biomass of ETL species increases. However, if there is a low threshold of host species needed to initiate or prolong an infectious event, then no amount of a particular IMTA species would be safe since a minimum biomass would need to be cultured for them to be useful to the IMTA farm. An important point to be considered in the evaluation of risk is the current situation. Although salmon farms are generally considered to be monoculture operations, they are actually incidental IMTA operations already. The ETL species that are being considered as IMTA candidates are fouling or wild species which already exist on site, albeit in lower densities. Salmon farms may have high densities of blue mussels that grow on the infrastructure and other benthic species such as sea urchins, sea cucumbers, and polychaete worms which exists in significant quantities on the benthos below the farms and in nearby habitats. These species have been present for over 30 years of salmon aquaculture development, and there is little published evidence that the presence of any species on the salmon farms have resulted in fish health concerns, although the dynamics between various cultured and wild species are beginning to be recognized (Salama and Rabe 2013). However, laboratory studies indicate the blue mussel (*Mytilus edulis*) is capable of destroying infectious salmon anemia (ISA) viruses (Skar and Mortensen 2007; Molloy et al. 2012) and several bacterial species such as *Escherichia coli*, *Micrococcus luteus*, *Bacillus cereus*, and *Renibacterium salmoninarum* (Birkbeck and McHenery 1982; Paclibare et al. 1994). Other viruses such as infectious pancreatic necrosis virus (IPNV) that accumulated in the faeces of the blue mussel were able to be passed on to naïve salmon smolts (Molloy et al. 2013). Similar results were found for the great scallop *Pecten maximus* (Mortensen 1993). Recent studies on the pathogenic cod (*Gadus morhua*) bacterium, *Vibrio anguillarum*, indicate mussels were able to remove the bacteria from the water column; however, bacteria encapsulated in their faeces were still viable and able to produce infections in naïve fish (Pietrak et al. 2012). The exposure of fish to re-suspended mussel faeces was artificially high in these studies and the authors acknowledged that the dynamics of faeces in natural situations may differ.

Bacteria also play another role in the dynamics of IMTA systems. Heterotrophic bacteria are major converters of organic carbon in the marine environment and are found ubiquitously in pelagic and benthic habitats. There can be significant conversion of particulate organic material into carbon dioxide and ammonia by this ETL (Carr and Morin 2002).

Parasites are another group that contribute to the dynamics of an IMTA site. Presently, one of the most destructive parasites for the Atlantic salmon farming industry is sea louse (*Lepeophtheirus salmonis* and *Caligus elongatus*). These two species cause extensive economic damage to the industry by significantly increasing the mortality rates of cultured salmon and have created substantial industry problems through the application of therapeutic chemical control measures, as well as management issues. Although IMTA practices are unlikely to exacerbate the problem with sea lice, there may be some benefits incurred as some studies now indicate several filter feeders are capable of ingesting larval stages of various invertebrate larvae (Lehane and Davenport 2002, 2004; Zeldis et al. 2004) including sea lice (Molloy et al.

2011; Bartsch et al. 2013; Webb et al. 2013). While IMTA filter feeders are unlikely to be the sole solution for the problem, they may be part of the control measures available for helping to control sea lice.

There are issues with other parasites that may be relevant to IMTA operations. Some parasites have an intermediate host before reaching their final species destination. Although there is very little information on the proposed IMTA species such as scallops, sea urchins, sea cucumbers, or polychaetes worms as being intermediate hosts to parasites of FTLs, it would not be advisable to grow large quantities of a potential intermediate host species in close proximity to the ultimate host, such as salmon, until preliminary screening trials are completed (Mike Beattie, Province of New Brunswick, pers. comm.). Additional research is recommended in regard to this aspect. This issues needs to be considered in terms of scale. Wild conspecifics of these extractive IMTA species are currently present on salmon aquaculture sites, and there has been no report of parasite infections exceeding normal levels that could be related to differences in densities in associated local invertebrates.

Larger organisms will also interact with the IMTA operations. Three-dimensional structure in marine environments creates additional habitat which is very attractive to settling organisms (Robinson et al. 2011). The infrastructure around aquaculture sites is substantial and is colonized relatively rapidly. Some of the organisms that have been observed to use aquaculture sites include American lobsters, blue mussels, sea urchins, sea cucumbers, juvenile herring, juvenile pollock, mackerel, sculpins, tunicates, hydroids, and several species of crabs. Some studies have suggested that salmon aquaculture sites not only act as fish aggregating devices (FADs), but can also act as a refuge areas where some species, including commercial ones, can increase their population numbers (Dempster et al. 2011).

Increasing the interactions within an IMTA site creates multiple linkages. The addition of biomass to the lease area will help with the conversion of particulate organic carbon into inorganic forms resulting in less cumulative loading to the benthos. The increased food energy available on the site may increase the recruitment rate of some species including commercial ones. Lobsters are regularly found on active salmon aquaculture sites (S. Robinson, personal observation) and any additional infrastructure associated with IMTA operations will likely increase this interaction. Juvenile herring (*Clupea harengus*) and pollock (*Pollachius virens*) regularly use the area beneath IMTA mussel rafts (S. Robinson, personal observation). Although no specific studies have been completed to date, it is presumed they are using the area for protection and feeding. If the IMTA structures could specifically be designed to enhance juvenile lobster (or other commercial species), beneficial linkages could be achieved between the aquaculture and wild harvest industries, similar to previous suggestions on sustainable coastal production systems (Newkirk 1996).

Interactions with wild species, however, may create problems through the transmission of diseases and parasites. It is anticipated that most IMTA species originate from hatcheries, with the exception of some species (e.g. mussels), that produce prolific qualities of pelagic larvae that are captured from the wild, and are initially disease and parasite free. The influx of wild species into the IMTA operations may result in the transfer of undesirable pathogenic organisms from the wild to the cultured species. Other wild-farmed species interactions may be through predation. Currently, eider ducks prey upon blue mussel and have been shown to impact unprotected populations of mussels growing on rafts on salmon sites. A flock of ducks can eat or detach over 20 tonnes of blue mussels from mussel rafts in days (S. Robinson, personal observation). As other species are introduced to the site, additional predators will likely be attracted to the high abundances of potential prey. Mechanisms will have to be in place to deal with those on a species by species basis.

## Summary

There are several factors that determine the efficiency of an open-water IMTA operation. Carefully selecting the proper species to meet requirements will ensure the appropriate trophic niches are filled and coexistence with the other species. The species should be easy to work with and produce enough value to augment revenue streams. The potential efficiency of the organism is another consideration for IMTA. Most organisms are relatively inefficient at converting food with some of the best ecological efficiencies of around 30%. Before a species is chosen, its physiology should be sufficiently understood so the grower is aware of expected transfer rates between trophic levels and the resultant waste production of that particular ETL so that they can better plan their operation. The nutrient waste production will depend on the total biomass cultivated, diet availability, and how efficiently the trophic linkages are established. The efficiency of those linkages strongly depends on the physical factors at the culture site, such as water depth, currents, and temperature. In addition, there will be a number of intentional and unintentional interactions within the ecosystem. These interactions can take the form of disease and parasitism at the small scale to predation events at the large scale. Understanding the scale and mechanisms of these ecological interactions will help to optimize outcomes and assist in the development new management policies for IMTA. The more efficient the IMTA system, the less nutrients (energy) will be available for transfer to the outside environment.

## DOES IMTA HELP REDUCE THE BENTHIC ORGANIC LOADING AT AN AQUACULTURE SITE?

Evaluating whether there is a net environmental benefit of IMTA to reduce benthic organic loading requires an understanding of the various processes that contribute to the benthic loading process. To assess this, the issue was broken into five sections:

- defining the loading environment,
- diet proportions,
- extraction capabilities,
- likelihood of this happening, and
- other environmental benefits.

### Defining the loading environment

There is a substantial amount of energy that is used in the industrial production of salmon. Early estimates (Folke and Kautsky 1989) suggest that it takes up to 720 GJ (gigajoules) to produce one ton of salmon, much of that from the natural ecosystem. The mass of fish food that is utilized to grow salmon is substantial considering the quantity of fish biomass that is being grown. However, fish are one of the most efficient animal converters of food in anthropogenic culture systems today and, therefore, are good choices for culture (Table 1). At peak biomass, salmon being fed at a rate of 1% (Handeland et al. 2008) of their body weight per day will produce over 2 tonnes of faeces with potential for bottom deposition unless converted by some other organism (bacteria or ETL species) (Table 5). It is important to note that this is an estimate of the maximum daily value of faecal deposition for a full site of 4.5 kg fish at warm temperatures, with radically different feed inputs compared to requirements for 60 g smolts introduced to cages in cooler spring temperatures at the start of the production cycle.

Salmonid diets are designed to be highly digestible and have been optimized over a number of decades to have low food conversion ratios (FCRs). An average FCR of 1.1 over the entire

production cycle is commonly reported by the industry. This means that 1.1 kg of dry food produces 1 kg of wet fish. It should be noted that these are not the same as trophic transfer efficiencies since the amount of water contained in the diet and the fish are not equivalent; thus, FCRs are generally only used operationally to monitor the efficiency of the feeding operation. However, an important point to IMTA processes is that the organic content of the diets are very high, resulting in higher levels of organic content in the faecal production. This feature is important when faecal production outputs are compared between different trophic levels.

A similar calculated approach was used to evaluate the potential organic loading rates of mussel culture on the benthic organic loading (Table 6). A total of 100 tonnes of blue mussels cultured on 4 commercial rafts located within the IMTA salmon farm would produce a total of 864 kg of faecal production per day. Comparatively, the salmon would produce 2,025 kg per day. However, unlike the salmon faeces, the organic content of blue mussel faeces is much lower resulting in approximately 20% of the organic output in comparison to salmon.

No estimates of faecal production rates are currently available for sea cucumbers or sea urchins because not all the rates have been measured in an IMTA setting. However, considering that the diets the animals are eating are similar to that of the mussels, and the volumes of culture might also be similar, one could reasonably expect that the outputs would be similar with regard to total and organic deposition. Studies on this aspect are currently underway.

Current velocities, particle densities, and water depth have important roles in determining the organic particulate loading rates to the bottom. There are important differences between the various types of organic waste, particularly in their settling velocities and composition. Different portions of an aquatic faecal load will settle at different rates. This is referred to as mass fraction settling velocities and these velocities can be assigned a statistical distribution. Settling of mussel faeces produced from ingestion of salmon farm organics and natural diets, settles 3 to ten times slower than salmon faeces and contains approximately half the organic content (Reid et al. 2009; Liutkus et al. 2012). The slower mass-fraction settling velocity and organic content of mussel faeces compared to salmon faeces means that an equivalent amount of mussel faeces would have a reduced potential for benthic impact. Certain thresholds of organic carbon deposition from aquaculture have been identified to be associated with adverse benthic impacts on soft bottoms (Cromey et al. 2002, 2012; Hargrave et al. 2008). A slower settling rate will disperse aquaculture faeces farther, reducing the carbon deposition per unit area. A reduced proportion of organic carbon per unit mass in mussel faeces should further reduce the area of benthic loading of organic carbon. Information on mass-fraction settling velocities and organic carbon composition of both faecal types were used to simulate a comparison of area benthic carbon loading between salmon and mussel culture. Data from several studies (Reid et al. 2009; Liutkus et al. 2012) and unpublished field studies were used to model the relative carbon deposition of mussel and salmon faeces along a hypothetical transect, as a means to compare the benthic impact potential between mussel and salmon faecal loads of the same mass, depth and current regimens (Appendix 2).

Modelling results indicate that, in 20 m of water, the loading rates to the benthos are substantially higher for salmon for both organic and inorganic components (Figure 3). The results also indicate the deposition of salmon faecal matter is substantially higher closer to the fish cages. Comparatively, mussel faeces settle farther away with lower amounts of organic and inorganic material. No data are currently available for other species, but similar calculations can be done once the biophysical properties of their faeces are known.

### Diet Proportions

Simply adding the various particulate organic contributions of the IMTA species to determine the net loading to the bottom of an IMTA site is insufficient since trophic interactions between

species are involved. The main underlying point of the IMTA concept is that synergies are created between species when they are grown in close proximity to each other. Thus, the waste that is produced from one trophic level becomes the inputs to another and the potential energy (or organic content) is reduced overall. This becomes significant when the higher organic content in salmon faecal particles is converted in the secondary ETL faecal pellets.

To understand the implications of the conversion of organic matter prior to bottom deposition, a model was developed to determine the dietary proportion of salmon culture organics that must be consumed by an IMTA mussel population before there is a net reduction in organic load.

Estimates of the net addition or removal of organic material by shellfish in IMTA systems can be made using absorption efficiencies of mixed diets (i.e. fish farm organic solids and natural diets) as a first step to estimate the proportion of the organic load from the farm that must be consumed by shellfish, before net organic load in an IMTA system is reduced (Reid et al. 2013). This approach initially circumvents the need for shellfish biomass and production data to provide first order estimates of mitigation potential. Once the proportion of salmon culture solids in the mussel diet is known, it is possible to ascribe production levels, deployment locations and husbandry practises required to achieve a desired decrease in the organic loading from the IMTA site.

Model development and equations are detailed in Appendix 1. A maximum threshold of 1/3 consumption (33%) of salmon solids available was applied to model simulations due to assumed particle size availability and delivery, discussed elsewhere in this document. The dietary proportion threshold (%) for the consumption of salmon solids necessary before the ETL becomes neutral in regard to organic loading is as follows: salmon faeces and high quality seston = 14.5%, salmon faeces and low quality seston = 19.6%, salmon feed and high quality seston = 11.5%, salmon feed and low quality seston = 15.6%. Changing the total particulate matter (TPM) consumed by mussels or the amount of salmon culture solids available did not change model outputs as long as the dietary proportions were maintained. To clarify this approach, an example of two model scenarios are shown below, where an IMTA site with a mussel population has decreased the organic load and one where the net organic load has been increased (Table 7). A net reduction in site-wide organic load will occur if mussels remove (absorb) more fish farm organics than organic material loaded in mussel faeces produced from indigestible seston.

The results of this simulation demonstrate that whether an IMTA extractive species contributes to the net organic loading at an aquaculture site depends on the proportion of the diet the species assimilates from other trophic levels. While both trophic levels are producing organic waste, the issue is whether the lower trophic levels are removing more organic material than they are adding.

Theoretically, the dietary demands of increasing mussel production could exceed the solids available from the farm, thereby imposing limits on the proportion of farm solids that could be consumed. Since additional farm solids cannot be consumed once they are fully utilized, any additional mussel production will result in increased organic loading by default, assuming the natural food sources are available. A substantial level of mussel production would be required for this to occur, and it is doubtful that this level of production would occur within the site lease area of a fully stocked commercial salmon farm in New Brunswick that is trying to minimize benthic impact. A more likely scenario is that the production levels of the extractive species would be grown in proportion to the calculated food levels available since revenues are generated from all IMTA components and the farm will be optimizing for all revenue streams.

## Extraction Capabilities

As long as the dietary proportion of salmon solids by an IMTA mussel population is maintained, the amount of TPM (natural diets and salmon culture solids) consumed will not affect the IMTA loading outcome, assuming minimal pseudo-faeces production. However, a number of factors can impose limits on the diet proportion of salmon culture solids that can be consumed by a mussel or other IMTA population. These include:

- size classes of salmon culture solids available,
- amount of salmon culture solids available compared to natural TPM concentrations,
- direction and depth of particle delivery, and
- frequency, duration and rates of delivery.

### *Size Classes of Salmon Culture Solids Available*

Although the total amount of solid organic waste originating from salmon feeding operations can be calculated and compared with data from the literature, there is much less information available on the size classes of particles present in the water column. This is an important IMTA concept as each ETL species has an optimal size range on which it can feed. If a particle is outside that range, the species will not be useful in dealing with that particle size class. For example, the accepted size range of food items in the blue mussel (*M. edulis*) ranges from approximately 3 to 100  $\mu\text{m}$ , with most of the diet less than 20  $\mu\text{m}$ . However, studies completed New Zealand (Zeldis et al. 2004) and the United Kingdom demonstrated that mussels could capture zooplankton up to 6 mm (e.g. Davenport et al. 2000; Lehane and Davenport 2002, 2004). In addition, ocean particles are not always single and may clump together in aggregates known as "marine snow" or "flocs". Mussels are capable of utilizing these flocs, which can attain sizes of 4-5 mm (Newell et al. 2005). The preferred size classes of food particles for sea cucumbers are unknown. Sea urchins, being grazing organisms, are capable of handling large food items measured in centimeters; however, information pertaining to the lower limit of the size range of their diet is unknown. Sea urchins can graze on epiphytic diatom films; however, the colonial diatoms are eaten in large pieces.

Measuring the size distribution of organic particles is difficult. Sizes can range from < 1  $\mu\text{m}$  (individual particles) to > 20,000  $\mu\text{m}$  (salmon food pellets and faecal casts) (Figure 4).

Instrumentation capable of measuring subsets of the size range, such as laser particle counters (LISST 100X, Sequoia Scientific, Inc. 2700 Richards Road, Suite 107, Bellevue, WA 98005), only work within the range of 2 to 300  $\mu\text{m}$  and the data are often complicated to interpret. *In situ*, high-resolution cameras are used for determining the abundance and size of flocs because the particles are so delicate. In many cases, particles are destroyed during the sampling process (e.g. Newell et al. 2005). Unfortunately, no one technique can measure all size classes simultaneously and provide information on temporal and spatial distribution patterns. Consequently, the proportion of loaded salmon culture solids within the filtering size range of blue mussels around the salmon farms is not known.

### *Amount of Salmon Culture Solids Available Compared to Natural Tpm Concentrations*

The closer IMTA organisms are to the point source of the nutrients, the higher the probability their diet will consist of aquaculture derived particles, although this partially depends on the settling velocity of the particles in the water column. The relative concentration of aquaculture organic particles or dissolved nutrients is highest at the source and becomes more dilute as they are advected away by currents and mix with the natural background material. There will also be a degree of patchiness in the natural seston concentration, as well as with the aquaculture-derived nutrients. In nature, the distribution of organisms is patchy, often being

determined by physical processes such as stratification or retention mechanisms. This can result in very high or very low concentrations of food that may not persist over relatively long periods of time. Different concentrations of natural food over time will affect the proportions of dietary food items in the diet of the IMTA species. Concentrations of aquaculture derived nutrients are also patchy since they are derived from the feeding operations in daily activities on the farm. Fish are fed during daylight hours (since the fish are visual feeders) and may occur in a single or multiple pulses depending on whether feeding is completed manually with blowers or delivered through pipes linked to feeding systems with camera feedbacks.

#### *Direction and Depth of Particle Delivery*

It is important for IMTA extractive species to be as close to the source of the nutrients as possible and to ensure they are in the waste stream to maximize their proportion of aquaculture derived material. This is sometimes difficult since structures of the aquaculture site often modify currents as water passes between, around, and under cage arrays. Adding additional infrastructure for the IMTA extractive species will further complicate this issue. It may be difficult to create a linkage between the nutrient source and the appropriate ETL for some species in the current configuration because of spatial separation. For example, to readily access sunlight for photosynthesis, seaweeds are generally grown in the top 2 to 3 m of the water column. If a portion of the inorganic nutrients are being released at the bottom of the fish cage (15 m), with minimal mixing and a mean current velocity of 10 cm/s, there is a decreased likelihood those nutrients will be available to the kelp for absorption depending on the distance between the seaweeds and the fish cage. In the case of seaweeds, there may be little difference between a nitrogen molecule originating from salmon or sea urchins and one originating from local wild species. For organic particles that can have near-field impacts on the benthos, having an efficient linkage between the different trophic levels is important. The development and engineering of IMTA sites are still in its infancy, but there is a role for marine engineers to optimize trophic linkages through infrastructure design of IMTA farming operations.

#### *Frequency and Rates of Delivery*

As previously described, currents can have a major effect on the delivery of nutrients to the IMTA species. Feeding on a salmon farm is generally sporadic with one or multiple feeding events during daylight hours which will generate organic particles. An additional pulse of organic particles may occur in the latter portion of the day as digested food is egested. During the pulse periods, the apparent concentration of aquaculture produce nutrients is high and may represent a high proportion of the particles that are ingested. However, during low periods, natural particles may predominate and subsequently represent a higher proportion of ingested particles. This is likely to occur at night, although there are little field data available to evaluate feeding of filter feeders or deposit feeders during these times. Current speed also influences the contact time of the nutrients with their respective extractive species at the appropriate trophic levels. As indicated above, if currents are too fast, pulses of food advected through the containment structures of the extractive species will be too rapid for the ETLs to efficiently extract aquaculture derived nutrients and they will be left to feed on the natural background material, thereby reducing their aquaculture-based diet proportions (Cranford et al. 2013).

#### **Evidence of Significant Feeding on Aquaculture Based Nutrients**

Laboratory studies report some shellfish species such as mussels and oysters can readily filter, digest, and retain fish food and/or faeces (Lefebvre et al. 2000; Redmond et al. 2010; Reid et al. 2010). However, evidence of the perceived growth benefit to shellfish in open-water IMTA systems has been contradictory in approximately 30% of the cases. Several studies report augmented shellfish growth or evidence of farm organic uptake in open-water IMTA systems (Wallace 1980; Stirling and Okumus 1995; Jones and Iwama 1991; Lander et al. 2004; Gao et

al. 2006; Peharda et al. 2007; Sarà et al. 2009; Jiang et al. 2012; Handå et al. 2012a, 2012b; Lander et al. 2012), while other studies reported no effect (Taylor et al. 1992; Parsons et al. 2002 (no controls); Cheshuk et al. 2003; Navarrete-Mier et al. 2010). This implies a substantial influence of site-specific factors that affect the concentrations and spatial distribution of finfish wastes through differences in nutrient supply (husbandry) and dilution (advection and dispersion) processes.

Determining the proportion of aquaculture derived organic nutrients in the diet of IMTA blue mussels or other ETL species on an ongoing basis is difficult. Ideally, the analysis of samples that have integrated a particular signal over time and the use of tracers such as fatty acids, stable isotopes or other variable can be used to infer the past diet of the animal. Currently, the tracers used in IMTA-related studies have been stable isotopes (Gao et al. 2006; Redmond et al. 2010; Jiang et al. 2012), fatty acids (Gao et al. 2006; Redmond et al. 2010; Olsen et al. 2012) and dietary pigments (Graydon et al. 2012). All of these studies show incorporation of aquaculture-related diets into somatic tissue. Two studies (Gao et al. 2006; Jiang et al. 2012) estimated that green-lipped mussels, *Perna viridis*, and Pacific oysters, *Crassostrea gigas*, could assimilate 30-35% of their diet from waste fish nutrients.

An alternative method to measure the amount of food incorporated into an IMTA species is the growth differential between animals grown in close proximity to aquaculture sites where they are more likely to inhabit the waste nutrient stream compared to animals grown further away which should have proportionally less aquaculture particles in their diet due to the effects of dilution. Any growth change is likely due to aquaculture-derived nutrients. Since the metabolic costs (carbon dioxide and ammonia production) and any reproductive products are not accounted for, animals have likely assimilated more food than the measured growth differential, thus under-estimating total assimilation rate.

In 2002, an experiment with small mussels held in experimental lengths of commercial mussel socking was conducted. After one year of growth the animals were sampled for the last time (Lander et al. 2012). In comparison to the reference site, blue mussels on the IMTA sites grew significantly larger in weight, particularly those closest to the cages. The growth differences in somatic tissue ranged from 30 to 109% larger (Figure 5). Within site comparisons indicated mussels closest to the fish cages grew 25% larger at the Atlantic Silver site and 40% larger at the Aquafish site (Lander et al. 2012), suggesting that the mussels closer to the fish cage were getting significantly more and/or better food.

In 2002, a second study used a tag recapture technique where individual mussels were labelled and suspended from either a cage or at a mooring 500 m away. Juveniles were suspended from June to November 2002 and this process was replicated at five different aquaculture sites (Figure 6). The results from this experiment corroborated the first one and demonstrated that mussels grown off cages grew substantially faster than those further away, supporting the concept of increased nutrient loads closer to the cages and the ability of mussels to utilize those resources for somatic tissue.

All growth trials conducted from 2003-2005 did not show significant differences between mussels grown near salmon cages compared to those grown in reference locations several hundred meters away. This was observed when sampling was done on entire rafts. Although the mussels all grew rapidly, no differences in mean weight were observed. These industrial-scale trials were often confounded by the locations available as reference sites and the natural thinning of the population that seemed to occur as larger mussels tended to fall off the lines in rough weather. Further work is needed at these commercial scales to determine the density dependent effect of large volumes of mussels grown in proximity to salmon cages.

### Other environmental benefits

Although not directly related to benthic organic loading, there are other environmental benefits that may accrue from using IMTA concepts. The first is the relative increase and intensification of the production that comes from an aquaculture site, both in biomass and economic revenues since new products are being produced. Assuming that protocols are developed and accepted into the working business model of the current aquaculture industry, IMTA meets part of the needs for the expansion of the industry. Virtually every business plan is premised on an annual rate of increase in order to compensate for changing revenues due to competition and the annual rate of inflation. In a monoculture industry, the only options the operator has are to increase the volume of production or to change the price of the product. Increasing the volumes of production often mean applying for further lease spaces and taking up more space in the marine coastal zone or potentially overloading the space they currently occupy. This creates a number of new socio-political issues with resource management. IMTA, up to a certain point, does not require additional area, but can generate increasing levels of revenue over the monoculture baseline, thereby fulfilling the need for the annual rate of increase in the business plan. Over time, this means less spatial area of natural ecosystems is required/desired to meet the coastal regions requirement for aquaculture production.

A second indirect environmental benefit to IMTA production is the potential synergies that are created in a multi-species environment. Although the potential positive and negative interactions of various species being grown in close proximity on a site were previously discussed, multi-species complexes are the norm in nature. Some of the most productive areas in the marine environment are reefs where a combination of three-dimensional structure and multiple trophic levels create an ecosystem where nutrients are constantly recycled on a regular basis. These systems persist for long periods of time because of their inherent stability due to biodiversity. There are some studies which would suggest that growing certain extractive species might be beneficial in helping to control some diseases and parasites which are now currently being controlled through the prescribed use of chemotherapeuticants (e.g. Skar and Mortensen 1997; Molloy et al. 2011; Bartsch et al. 2013; Webb et al. 2013). If the use of IMTA species could reduce the frequency needed to control the aquaculture pests, then there would be a net environmental benefit by not having these chemicals in the water impacting wild species and developing drug resistance in the pathogenic ones. An example of this control of diseases can be found in the shrimp industry. In experimental pond trials, green mussels, brown mussels and oysters grown with shrimp significantly lowered the concentrations of luminous bacteria and increased the survival rates of the shrimp (Tendencia 2007). This IMTA system is being considered for scale up into ponds to test this in the field situation. If successful, it could be another approach in the fish health management of shrimp culture.

### Summary

Determining a net environmental benefit for IMTA requires an understanding of the loading environment and the scales involved. Growing large numbers of FTLs, such as salmon, for commodity markets requires large amounts of food which results in large amounts of organic and inorganic waste. The ETLs which feed on the organic waste nutrients (either natural or aquaculture derived) produce their own levels of waste, which can be significant. However, the organic content of the waste from the ETL can be significantly lower compared to the original source, which makes a large difference to the loading of organic carbon to the bottom. Therefore, understanding the organic carbon available and its mechanisms of delivery will determine the amount of food available. Whether or not an IMTA ETL organism contributes to the net organic deposition to the sea bottom from the IMTA site depends on the proportion of its diet obtained from an FTL. Consumption of a diet results in the conversion of much of the

organic carbon into carbon dioxide which dissolves into the water and drifts away from the site to be recycled by primary producers. The remaining organic carbon in the diet that is not assimilated is egested as faeces, some of which will settle to the bottom within the site lease area. Estimates from this study indicate that diets of ETLs must consume 10 to 20% food from the FTL's (i.e. salmon culture solids) before there is a decrease in the net organic loading at an IMTA site. Whether an ETL can accommodate this amount of diet depends on the size classes of diet available and the ability to exploit it. The ingestion potential of an organism is primarily dependant on its morphological structures and the contact rates with the food. Several studies in the literature have shown that some open-water extractive species can utilize fish farm waste nutrients at a level of approximately 30% of their diet. To date, empirical field data on the blue mussel suggests these animals are capable of assimilating similar amounts of fish-derived nutrients in their diets as long as they are in contact with the farm derived food. The organic content of the waste from the ETL will be significantly lower compared to the original source, which makes a large difference to the loading of organic carbon to the bottom. Mussel faeces settle at a much slower rate and contain much less carbon than salmon faeces, depositing less carbon per unit area, and consequently resulting in a lower relative benthic impact. However, it is considered very unlikely that IMTA, using only fine particulate feeders (mussels) in SWNB, has significantly reduced loading of total particulate matter to the benthos at the sites.

## **DOES THE CURRENT SCALE OF IMTA IN SOUTHWEST NEW BRUNSWICK PROVIDE ANY BENEFITS?**

### **Introduction**

There is a great deal of interest in the potential benefits that might accrue from using IMTA practices such as what operational scale (i.e. spatial scale), species mix, and what biophysical attributes, would be required to result in a net environmental benefit (e.g. a reduction in the rate of carbon deposition). This section projects some of the presently available information on the development of the IMTA system into the future. Since work has only been done on two of the extractive trophic levels (ETLs - the inorganic extractive level and the fine particulate extractive level), many of the subsequent analyses are based on best estimate scenarios taken from existing research, as well as information gathered from the literature. The section is subdivided into:

- assessing the benefits,
  - nutrients
  - general categories
- present-day status of IMTA, and
- future configurations.

### **Assessing the Benefits**

In order to assess whether the IMTA concept is beneficial to the logistic and sustainable operation of the aquaculture industry and to environmental management from the perspective of various government departments, the net balance of the costs and benefits that are associated with the practice on both an economic and social basis must be determined. A comparison is available in Table 8.

### *The Multiple Roles of Nutrients*

Ecosystem health and consequently, biodiversity, are influenced by nutrient concentrations in aquatic systems since concentrations of limiting nutrients (typically nitrogen in marine systems and phosphorus in freshwater) frequently dictate the level of primary productivity. High nutrient concentrations may result in excessive primary productivity (eutrophication), where resultant algal blooms ultimately die, sink and strip oxygen from the water column upon decomposition. In extreme cases, the decomposition of organic material, whether from algae or direct anthropogenic sources (e.g. sewage) will cause so called, "dead zones" (Dodds 2006). "Dead zones" have been a common re-occurring problem during the last half century, linked largely to the delivery of reactive nitrogen and terrestrial phosphorus to the oceans, which has increased threefold from pre-industrial agricultural times (Díaz et al. 2009). It is these negative impacts of excessive nutrients that are so often associated with nutrients in general. However, low concentrations of nutrients may also affect ecosystem status, resulting in oligotrophic environments, with similarly low productivity and limited food sources. In fact, in some aquatic environments, such as freshwater highlands, removal of nutrients is a significant concern, and methods to recycle phosphorus in a carefully controlled and ecologically sensitive way to restore sufficient fisheries production level, have been advocated (Stockner et al. 2000). Herein lays the paradox of nutrients. Too much or too little is "bad", and arguably the relationship between nutrient concentrations and ecosystem health is a quantity issue more than a substance issue.

One of the biggest challenges for quantifying effects to ecosystem health is the consideration of scale and this is also true of nutrient availability. Most ecosystem services are delivered at the local scale, but their supply is influenced by regional or global-scale processes and frequently, benefits accrued at one scale may result in costs at another (Carpenter et al. 2006). Ocean eutrophication from run-off of agriculture fertilizer (Beman et al. 2005) is arguably an example of this. Ironically, fertilizer is added to augment nutrient levels insufficient in the soil as a means to increase crop productivity. In many parts of the world, this results in large nutrient imbalances (more nutrient is added than removed within crops, e.g. USA, China), with the 'extra' nutrient run-off directed to surrounding waters (Vitousek et al. 2009). Issues of scale, transference of costs to other regions, and sensitivity (i.e. trophic level) of the receiving environment require consideration.

The "nutrient paradox" may also be manifested at the management level. Should policies be applied to relevant scales? Should they be applied at the source of an impact, the receiving body or both? In the case of nutrients, where addition or removal can be either positive or negative depending on the type of environment and scale of effects, over-arching policies may be insufficient. For example, the application of nutrient trading credits (NTC) as an incentive to removal/reduce nutrients to mitigate or prevent eutrophication may have merit (e.g. Lindahl and Kollberg 2009). NTCs would not be necessary in environments where moderate enrichment stimulates productivity and biodiversity. Therefore, the degree of nutrient removal should be a function of optimizing ecosystem performance at the scale or environment of interest. Obviously, there is some anthropogenic interpretation on what "environmental performance" means.

Organic loading to the benthos from aquaculture produces similar effects to organic loading from other sources. As organic enrichment on soft bottoms increases, biota typically transitions from a high biodiversity, to a greater abundance of more "pollution" tolerant species (Pearson and Rosenberg 1978) until anoxia ultimately occurs. A summary of the general effects of excessive benthic marine organic enrichment is illustrated with a nomogram by Hargrave et al. (2008). Increased organic carbon flux to the benthos is ultimately accompanied by increases in total free sulfides with corresponding decreases in oxygen, pH, Redox potential ( $Eh_{NHE}$ ), and biodiversity. The Shannon-Weiner Index, Hurlbert's Index, Infaunal Trophic Index, mean number

of taxa, and number of arthropod classes all show a decrease in biodiversity associated with increased organic carbon flux. It is important to note however, that this transition illustrates resultant effects of increasing sulfides and decreasing oxygen as a response to organic flux. Organic flux that is 'excessive', results in oxygen decrease and increase in sulfides since the rate of organic carbon input exceeds the rate of loss. Under some conditions of moderate enrichment, increased biodiversity may also accompany high abundance. Pearson and Rosenberg (1978) refer to this as the ecotone point or transitional zone. However, while this zone may be spatially limited, it may explain why, biodiversity may be increased with greater organic flux relative to background levels.

Numerous aquaculture studies on the benthic effects of excessive organic deposition have been completed. A limited survey of this literature yields conflicting results. A number of studies report measures of benthic biodiversity near aquaculture sites has increased relative to background levels (Macleod et al. 2006; Brooks and Mahnken 2003; Kutt et al. 2007; Brooks 2001; Karakassis et al. 2000; Borja et al. 2009; Apostolaki et al. 2007; Dimech et al. 2002; Kempf et al. 2002; Papageorgiou et al. 2009). Other studies have reported some measures of increased species abundance and/or biomass (typically pollution tolerant species), but not necessarily increases in biodiversity (Felsing et al. 2005; Karakassis et al. 2000; Klaoudatos et al. 2006; Brooks et al. 2003; Edgar et al. 2005; Felsing et al. 2005; Borja et al. 2009; Vidovic et al. 2009; Dimitriadis and Koutsoubas 2008; Nickell et al. 2009). Still others report either a decrease or no increases of biodiversity (Grego et al. 2009; Gao et al. 2005; Tomassetti et al. 2009; Fabi et al. 2009; Danovaro et al. 2004; Nickell et al. 2009; Felsing et al. 2005; Aguado-Giménez et al. 2007; Vidovic et al. 2009). These aforementioned studies had a variety of objectives, occurred in a variety of environments and consequently, employed a variety of sampling protocols. Direct comparison between them is not possible. However, where a depositional gradient of organic material is not always excessive and does not promote anoxia or hydrogen sulphide generation, a pattern appears to emerge. Large increases of infauna abundance and biomass at the local scale are frequently seen, with highly elevated levels of species richness in the transitional zone (Kutt et al. 2007; McKinsey et al. 2006a). This suggests that generalizations regarding the effect of organic loading from aquaculture are not reflective of the diversity of environmental and spatial responses. Generally, in the Bay of Fundy and elsewhere, enrichment effects in the area surrounding the salmon cages are observed within 50-100 m (Stucchi et al. 2005; Chang et al. 2007) and where organic carbon loading rates are greater than 2 g C/m<sup>2</sup>/d (Hargrave et al. 2008).

### Present-Day Status of IMTA

The current state of development of IMTA in SWNB is best described as 'pilot scale'. To date, only two ETLs (inorganic extractive, pelagic extractive) have been investigated, although there are currently several studies on benthic species underway, e.g. the Canadian Integrated Multi-Trophic Aquaculture Network (CIMTAN). Development of IMTA has primarily occurred within the Back Bay and Bliss Harbour areas outside of Passamaquoddy Bay. The main sites that have been occupied are Crow Island (MF-37), Charlie Cove (MF-276), Man-O-War (MF-32), Hills Island (MF-26), and Aquaventures (MF-30). Experiments have also been completed around Fairhaven (MF-59) and St. Andrews (MF-377) sites (Figure 7). Orientations of fish cages were unique to each site so there was no uniformity in the cage arrays or number of cages. IMTA structures for blue mussels and seaweed were generally located where it was convenient for the operation of the salmon farm and space was available. This is not surprising since salmon farms were established, active, working sites at the initiation of pilot scale work, and the IMTA research portion was not permitted to interfere with the day-to-day operations of the business. Despite efforts to do so, rafts were not always well-located in the down-stream effluent plumes coming from the salmon farming operation. Generally, for the blue mussel, there were two to six

rafts on each aquaculture lease. Dimensions of the rafts changed over time as modifications were done to make the work more efficient. The current design is a series of concentric circles stocked with mussels on a continuous longline system. Depending on the design, the capacity of the raft could be between 20 and 50 tonnes at peak production, but most rafts were not stocked at maximum densities. Therefore, it is not possible to easily calculate the biomass for each raft. However, if each raft produced 20 tonnes of blue mussels (20 t = 1.63 million mussels filtering 4 L/h of salmon-derived seston at a concentration of 3 mg/L 50% of the time due to changes in the tidal direction), each raft would consume approximately 237 kg of food per day at peak production, assuming it was in the right position to capture the food. Considering the placement of the rafts, it is improbable that the mussels significantly lowered the benthic loading from the salmon farms. For a typical farm of 300,000 fish producing 1559 kg of organic nutrients daily (mean salmon weight 4.5 kg, feeding at 1% body weight per day and producing 15% faeces), this would equate to about 15% of the total salmon output. Economically, at a farm gate price of \$0.65 per pound, each raft would have a gross value of approximately \$29,000.

### Future Configurations

In the future, the design of the IMTA aquaculture sites will have to evolve both structurally and in complexity as the new sites will have to accommodate additional species and ensure that the flow of water is sufficient for the salmon, as well as efficiently connecting the various trophic levels. While the site requires more three-dimensional space for the benthic component, it may not be necessary to expand the boundaries of the aquaculture lease if enough product can be grown in the existing space. If the area of the existing IMTA aquaculture leases just outside the boundaries of the outermost cages is measured, there is a substantial amount of lease space that can be used for subsurface IMTA structures (Table 8). On average, there is approximately 77,000 m<sup>2</sup> within the cage edge boundaries, although there would be logistic issues working underneath the existing cage designs. If 3 additional IMTA extractive species were grown on the sea bed, using only 10% of that area, this would result in an area for each species of approximately 2,500 m<sup>2</sup>.

The construction and organization of future IMTA sites will have to evolve to optimize several aspects, which include:

- Ensuring that each of the structures in the IMTA site are easily accessible by farm workers for installation, stocking, maintenance and harvesting.
- Optimizing the flow field around IMTA sites ensuring currents are fast enough to deliver oxygen and food to where they are needed, but slow enough, in some sections, so contact rates between the FTL and the ETL are sufficient to maximize extraction.
- Structures that build on the synergy already existing within the aquaculture operation. Materials should be similar in some of the basic elements, such as grid systems, cage materials and harvesting equipment.
- The development of specialized teams to handle different aspects of the operation since planning will need to consider the natural life history requirements of multiple species.

A possible configuration of future IMTA sites has fish cages around the periphery of the lease with extractive species in the middle and to the sides (Figure 8). This example uses a combination of 14 salmon cages (F) containing a total of 300,000 fish with 4 blue mussel rafts (M) and 16 engineered containers for benthic or mid-water extractive species (S). Seaweeds would be located further upstream or downstream of the highest densities of fine organic particles being released from the site, which settle on the blades of the kelp creating quality problems after drying.

If data on the spatial area potentially available to each ETL on existing salmon aquaculture lease were used to project culture densities along with feeding rates and relative values the benefits to production could be evaluated. It is evident in Table 10 that substantial quantities of organisms can be grown on the hypothetical farm within the existing lease space. Each species could be grown on 4% or less of the bottom lease space around the cages, suggesting that none of the structures have to be directly underneath the cages or outside of the lease boundaries creating issues with regulatory approval or with health and safety concerns for divers operating in the area. Feeding rates were projected at 0.75% body weight per day. For sea urchins, this was a reasonable estimate based on lab trials (S. Robinson, personal observation), but is more of a general estimate for the other species. Consumption values are predicted at peak biomass; however, when totalled, assuming the three species are capable of achieving reasonable contact rates with particulate nutrient streams from fish, the potential scales of consumption (1,694 kg per day) generally match the salmon nutrient output at peak production (2,025 kg per day from Table 5). While this is speculative, it does show that a significant reduction of organic waste to the sea bottom is theoretically possible.

One issue resulting from this approach is the source of juveniles for culture. While some juveniles may be collected using traditional spat collection techniques (e.g. scallop spat bags) from some prolific spawners (e.g. mussels and scallops), other species will have to be produced from hatcheries. The numbers involved would be too high for natural collection and the ecological impacts from harvesting benthic juveniles would be unacceptable. Much of the hatchery technology for some of the proposed IMTA species has already been developed, particularly in Asia.

### **Summary**

Adopting an IMTA philosophy for aquaculture production results in a number of costs and benefits. Costs may include increased nutrient loading, complexity in infrastructure and knowledge requirement for operators, and potential increases in the number of species interactions that may affect fish health. Benefits may include proactive methods of dealing with nutrient loading, increased revenue streams, product diversification, and reduction of some fish health issues. In SWNB, the present IMTA configurations are still in development, and, while their current configurations have likely not significantly lowered benthic loading at salmon farms, mussels have been used to remove fine particulates generated by salmon and kelp have extracted some fraction of soluble inorganic nutrients resulting from salmon metabolic and respiratory processes. Sea urchins and other species have the potential to be more effective at reducing benthic organic deposition beneath salmon farms. In the future, the design of the IMTA aquaculture sites will have to evolve both structurally and in complexity as the new sites will have to accommodate additional species and ensure that the flow of water is sufficient for the salmon, as well as efficiently connecting the various trophic levels. Calculations of the area available on existing aquaculture sites suggest sufficient space is presently available on most existing leases to accommodate new IMTA modules. Projections on potential production volumes suggest that significant quantities of organisms could be grown and are theoretically capable of assimilating much of the fish waste nutrients (assuming appropriate contact rates). The production of many of the new ETL IMTA species will tie closely to hatchery production due to the volumes required, timing, and the potential impact of harvesting juveniles from the wild.

### **AT WHAT SCALE WOULD IMTA HAVE MEASURABLE IMPACTS ON OTHER ASPECTS OF THE ECOSYSTEM?**

Ecosystem changes resulting from aquaculture could occur on a number of scales and trophic levels. Numerous reviews on ecosystem impacts related to aquaculture have been written. The

CSAS review (Anderson et al. 2006) on cumulative and far field fish habitat effects examined many of the factors that related to ecosystem-scale effects for shellfish aquaculture such as the importance of scaling analyses, cumulative effects of shellfish aquaculture such as grazing and biodeposition, as well as some case studies. A second CSAS review (McKinsey et al. 2006b) extensively summarized the effects of shellfish aquaculture on fish habitat. This review included aspects of ecological functions on filtration, biodeposition, nutrient cycling and habitat. These processes assessed the potential pelagic and bottom impacts of shellfish aquaculture. A third CSAS review (Cranford et al. 2006) summarized the indicators and thresholds for use in assessing shellfish aquaculture. Much of the information contained in these reviews is directly relevant to all of the processes that are occurring within the IMTA concept since many of the species and techniques being used for IMTA ETLs are those that are already being used in culture. Therefore, literature that has been summarized in these CSAS documents will not be further reviewed.

The focus will be specifically on industry-generated questions on changes resulting from IMTA practices that could potentially occur in the ecosystem related to changes in phytoplankton and intertidal species such as the softshell clam. Based on the literature and projections based on data from this project, the following framework will be used:

- Methods for detecting ecosystem changes
  - general trends in the Western world
- Phytoplankton interactions
  - addition
  - removal
- Secondary production interactions
  - intertidal species such as clams
  - creation of habitat
  - predator interactions

### **Methods for detecting ecosystem changes**

Understanding ecosystem changes within the Passamaquoddy Bay area resulting from the contribution of IMTA species on commercial salmon aquaculture sites is a formidable challenge. The scientific community does not fully understand the current stable state of the local ecosystem or which suite of baseline metrics should be used during an assessment. Potential upcoming temporal changes due to factors such as global climate change, sea level rise, and ocean acidification make it even more complicated. Compounding the issue is that any ecosystem impact empirically studied from IMTA practices is undoubtedly partially confounded by effects from other activities using the coastal zone such as other forms of aquaculture (e.g. salmon), fishing, coastal development, housing, fish processing, and primary industries such as logging and pulp and paper. These impacts would be present at far-field scales, meaning distances away from an aquaculture farm in the range of a kilometre to tens of kilometres.

Despite the complexity of the system, there are basic concepts that could be used to detect changes over time. One of these concepts would be the examination of habitats for major shifts in populations that are more dramatic than those due to inter-annual variability. In the ecological literature, populations of organisms are known to live in reasonable stable states which are often known as "multiple stable equilibria". Effectively, this theory states that ecosystems can exist in several different forms and that they have a tendency to remain in any one particular state unless they receive a large enough perturbation to move them to another, at which point they remain stable at that new state until they are moved again by some stimulus (Petraitis and Dudgeon 2004).

The multiple stable equilibria concept has been used in the field of marine ecology in the past in the study of coral reefs and subtidal kelp beds. For example, kelp forests and coralline algal barrens on the west coast of North America have been shown to be two stable states that can occur. Healthy kelp beds can withstand the grazing pressures from sea urchins and maintain their status for long periods of time. However, if some stressor affects the ability of kelp beds to withstand the grazing pressure of sea urchins, the kelp beds will transform into coralline algal barrens where grazing pressures by sea urchins will maintain the ecosystem in that state for long periods of time as well (Petraitis and Dudgeon 2004). In a second example, coral communities in the Caribbean can exist as long as there is grazing pressure to reduce macroalgal densities. If there is a shift in the controlling factor of grazers, macroalgae can grow and dominate the community by instituting changes in structure that make it difficult for grazers to re-establish their presence. Again, these two stable states can exist for long periods of time (Mumby 2009). A third example demonstrates a shift in processes that can occur in an ecosystem that creates two different states, this one from suspected human intervention. In Chesapeake Bay the industrial harvest of the American oyster occurred in the early 1900s. Continually increasing fishing pressure was applied to the oyster reefs in the bay as demand for the product grew (Newell 1988). The author estimated that in the early 1900s, the oyster population of Chesapeake Bay could filter 80% of the entire top 9 m of the water column of the bay per day. Due to stressors on the system from excess harvesting and disease, this filtration was reduced to less than 1% in the late 1980s. In addition to the reduction of filtering, there was an increase in the amount of nutrients available due to human population growth. The result in the ecosystem was a shift in the production from a benthic dominated community with several commercial species to a pelagic dominated one characterized by an abundance of phytoplankton and zooplankton. This has remained the case for several decades, despite efforts to restore the filtering capability of Chesapeake Bay and reinitiate the previous commercial harvests.

### **Phytoplankton Addition**

Some of the general examples noted above on multiple stable equilibria show that large systemic changes are evident at some of the key trophic levels. For example, changes in filtering capacity and nutrient addition in Chesapeake Bay resulted in changes in the primary producer trophic level with phytoplankton and also subsequent changes in the secondary production levels with zooplankton (Newell 1988).

Examining this concept for IMTA for the Passamaquoddy Bay area, there will be a certain amount of inorganic loading from the feeding activities of the salmon and various invertebrate species such as the mussels, sea cucumbers, sea urchins, etc., through the production of ammonia and carbon dioxide via metabolic processes. Salmon have previously been shown to be major contributors to the nitrogen nutrient pools in the water column of the Limekiln area (Strain and Hargrave 2005). If nutrients were limiting in the system, and there was an increase in the nitrates closer to the salmon farms, an increase in the chlorophyll levels in the area might be expected and possibly the appearance of algal blooms, toxic or non-toxic. However, the existing data do not support this concept. Nitrate levels measured in the water column in areas of high salmon farming versus offshore were no different (Martin et al. 2006; Strain et al. 1995). The large volumes of water entering into Passamaquoddy Bay twice a day ( $1-2 \times 10^9 \text{ m}^3$ ) may contribute to these findings (Trites and Macgregor 1962). A review of the studies on the correlations between aquaculture and harmful algal blooms internationally concluded there was no evidence to suggest any cause-effect relationship (Cranford et al. 2006). This implies that the level of nutrients being released by the salmon either have no detectable effect on the toxic phytoplankton populations or the effect is operating at different spatial scales than the monitoring can detect. Additionally, a review of nutrients and primary production in the region

concludes that the primary production in SWNB is light limited rather than nutrient limited (Harrison et al. 2005).

Empirical studies on IMTA aquaculture sites examining seston concentrations and chlorophyll<sub>a</sub> levels upstream and downstream of salmon farms indicated significant increases in seston levels in close proximity to the farm cages, but no significant difference in the chlorophyll levels upstream or downstream or within the cage array suggesting that the chlorophyll levels in the area were all at background levels (Lander et al. 2012). However, data from a study completed on *Mytilus galloprovincialis* mussels in the southern Mediterranean near Sicily indicated there was increased seston available and increased levels of chlorophyll in close proximity to the fish farm cages due to the increased level of nutrients available (Sarà et al. 2009). In the presence of optimal conditions and limiting nutrients, changes in the phytoplankton community, with nutrients released from fish farms, are possible. Given the degree of flushing in the Bay of Fundy, inorganic nutrients will likely be re-cycled away from the actual source.

### **Phytoplankton Depletion**

In addition to contributing nutrients that may enhance phytoplankton growth, it is plausible that the installation of large quantities of filter feeders, such as blue mussels, sea scallops or sea cucumbers might reduce the phytoplankton populations, or subsets of them, if the culture densities get high enough. Phytoplankton depletion has been observed where high culture densities of filter feeders are grown (see review in McKinsey et al. 2006b; Cranford et al. 2006). Commercial aquaculture sites growing blue mussel in bays in Prince Edward Island were shown to significantly reduce the phytoplankton populations resulting in lower growth rates of the mussels on the farms (Cranford et al. 2006; Grant et al. 2008). In addition, phytoplankton remaining in the farmed areas are often in the pico-phytoplankton size classes which are too small for some filter feeders to effectively exploit (Cranford et al. 2006).

It is questionable whether there would be an effect on phytoplankton levels in Passamaquoddy Bay and the outside islands with the addition of ETL filter feeders on IMTA sites. For example, over the last 60 years, there have been significant reductions in soft- shell clam populations, one of the dominant intertidal filter feeding species in the area. In the 1950s, the average landings of clams were approximately 3,000 tonnes. If that harvest represented 10% of the biomass, then would be a standing stock of 30,000 tonnes. From 1989 to 2008, average landings for clams were 403 tonnes. Assuming this represented 10% of the biomass, we would have a standing stock of 4,030 tonnes or a difference of 25,970 tonnes of clams that are missing from the ecosystem. Assuming that the average size of clam weighs 16.5 g and each clam filters 2.5 L per hour (Joergensen and Riisgaard 1988), this results in the loss of filtering of water by soft-shell clams in the intertidal zones of approximately 95,000,000 m<sup>3</sup> of water per day. Comparatively, if there were 50 IMTA farms, growing four rafts of blue mussels each, with 5 million mussels on each raft, with each mussel filtering 4 L per hour, those mussels would filter 4,000,000 m<sup>3</sup> of water per day or 4.2% of what the lost soft-shell clams used to do.

Undoubtedly, in the natural environment, other organisms have utilized the seston that the soft- shell clams used to use. However, the scales associated with the addition of mussels as one of the IMTA organisms suggest mussels would cause few problems with the depletion of phytoplankton, except perhaps at a very local level around the farms as has been experienced in other mussel farming operations regionally and internationally. Addition of scallops is also likely to result in minimal impact to existing phytoplankton populations.

### **Secondary Production**

Effects of IMTA operations on intertidal species, such as the soft-shell clam, may occur in various ways. First, the release of nutrients from the IMTA site may create unintended consequences. Robinson et al. (2005) indicated there was a possibility of nutrients from a

salmon farm reaching a beach and stimulating the growth of the green alga *Ulva* sp. The uncontrolled growth of *Ulva* sp. covered the clam flats, led to the displacement of soft-shell clams, resulting in economic losses for clam fishers. This example was found to be unique since the salmon farm was located in the mouth of a small bay with minimal flushing. Fish farms in close proximity to additional beaches did not experience similar algal blooms. It can be concluded that, under optimal condition, beaches can be affected from salmon aquaculture operations if the waste nutrients are not controlled. Similar concerns were raised in the west coast in British Columbia (Heaslip 2008). Using evidence based on traditional knowledge, concerns were raised that salmon farming was increasing the levels of green seaweed (*Ulva* sp.) on intertidal beaches beyond historical levels. In this study, there were suggestions that changes in the sediment characteristics may have resulted from the salmon farming operations. Similar comments have been expressed occasionally by local clam fishers in the Bay of Fundy regarding changes in sediment composition on intertidal clam beaches. Results from one study that analyzed the soft mud on the shore found no traces of aquaculture food pigments (canthaxanthin) in the samples that could trace the origin of the organic sediment back to the aquaculture industry (Zitko and Robinson 1996). More work on the frequency of occurrence and cause-effect should be done on the far-field effects of anthropogenic-derived nutrients in the natural environment.

Increasing the number of IMTA extractive species will likely increase the reproductive rate of a number of the wild populations in the Passamaquoddy Bay/Fundy Iles since the IMTA animals are endemic to the region. Recently, clam fishers have observed increasing numbers of juvenile mussels settling on the subtidal areas of a few beaches. It is questioned whether this increase has resulted from cultured mussels being grown on the IMTA sites. There is a strong likelihood that animals contained within the IMTA sites will spawn since they will experience one or more spawning seasons prior to harvesting. The question is whether the resulting larvae from the IMTA organisms will have a higher survival rate than their wild conspecifics. Early mortality rates are high in many marine animals that are prolific spawners and the larval periods for many species include a planktonic stage in which they will drift for four to six weeks. Depending on where the larvae are in the Bay of Fundy, they could move substantial distances away from their farm origin during that time. Trying to determine what population they came from would be challenging, but some genetic methods or some elemental tracers might help.

Understanding the recruitment issue regarding the contribution of the IMTA larvae compared to the wild larvae revolves around the mixture of larvae from IMTA sites and those from their wild conspecifics. There is approximately 730 km of shoreline in the Passamaquoddy/Fundy Iles area. Assuming the average stretch of shoreline supports a mussel population of 500 animals per linear meter (from high tide to low tide), a population size of approximately 365 million mussels is estimated to be present. If 20 IMTA farms each raised 4 rafts of cultured mussels the total number of mussels would be 90 million, or approximately 25% of the assumed wild population. While it is a significant proportion, it is probably not enough to dominate the wild population or change settlement patterns on beaches. However, without further sampling and analysis, it is impossible to extrapolate accurately any further.

### **Creation of Habitat**

When aquaculture structures are placed in the water, the three-dimensional habitat is colonized by a variety species. Several species have been observed to occupy the IMTA space to gain either food or protection from predators. This is not a unique feature. McKinsey et al. (2006b, 2011) highlighted several studies that indicated mussel farms create substantial amounts of biodiversity on the growing structures, which generally exceed that which might be lost on the bottom due to the organic loading characteristics of the farm. The role of three-dimensional structure is anticipated to play a major role in the benthic component of the IMTA systems

(Robinson et al. 2011). Studies on the utilization of the infrastructure of salmon farms by wild fish species in Norway concluded that fish farms were acting as fish aggregating devices (FADs), as well as nursery areas and a refuge for some wild commercial species such as cod and haddock (Dempster et al. 2005; Dempster et al. 2009; Dempster et al. 2011). The use of aquaculture sites by wild species is still poorly understood.

### Predators

Once a food source is available in high densities, it will attract organisms that use them as prey items. This is a common feature with culture systems (aquatic and terrestrial). For the salmon industry, seals, sea lions, other fish, predatory birds, and marine mammals such as otters or mink, can create problems with the production cycle, particularly on the smaller fish. For the traditional shellfish industry such as cultured mussels or oysters, predators such as diving ducks (Dionne et al. 2006), otters, or some invertebrate predators such as carnivorous gastropods or parasites such as polychaetes (*Polydora* spp.) can be problematic.

### Summary

There is the potential for IMTA operations to contribute to ecosystem stressors via nutrient waste addition and, in some scenarios, localized species addition. However, detecting small changes due to the IMTA beyond that of salmon aquaculture will be difficult. It is more important to monitor for larger scale changes to the ecosystem that could drive a new steady-state system, which may not be as compatible for human use. Although IMTA trophic levels in general may add increased nutrients at the local scales, there is no evidence in SWNB of nutrient limitation or eutrophication due to dissolved nutrients occurring except at very local transitory levels. This may be a result of the large volumes of water exchange from the Bay of Fundy into the coastal area and is supported by other studies. It is not expected there will be many changes to the phytoplankton on a broad scale, although there may be some local depletion levels around the farms due to filter feeders. The effects on secondary productivity through interactions with intertidal species are less certain. Depending on the hydrographic conditions of the intertidal zone, there could be interactions with the increased nutrients and epiphytic algae that create negative consequences for intertidal organisms. More research on larger ecosystem-scale effects is warranted.

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## TABLES

Table 1. Comparative table of trophic transfer efficiencies of various organisms based on food ingested to biomass gained.

Organism	Trophic Transfer Efficiency	Reference
Salmon	36%	this paper
Chicken (grain)	30%	Lindroth 1993
Pigs (grain)	11%	Lindroth 1993
Beef (grain)	5%	Lindroth 1993
Beef (grass)	3%	Lindroth 1993
Giant silk moth	10-15%	Lindroth 1993
Plankton	20-27%	Gaedke and Straile 1994

Table 2. Estimation of faecal composition and the amount of faeces produced from the consumption of a typical Atlantic salmon feed<sup>1</sup> for grow out sized (>2000 g) fish.

Variable	Proximate composition of dry salmon feed (% <sup>2</sup> )	Digestibility <sup>3</sup> (%)	Amount Digested (%)	Amount in faeces (%)
Protein (min)	39	90	35.1	3.9 <sup>4</sup>
Fat (min)	33	95	31.4	1.7
NFE <sup>5</sup> (max)	10	60	6.0	4.0
Fibre (max)	1.5	10	0.15	1.35
Phosphorus (approx. <sup>6</sup> )	1.2	50	0.6	0.6
Minerals <sup>7</sup> (max)	6.8	50	3.4	3.4
Moisture (max)	8.5			
Total	100			14.9

<sup>1</sup> Optiline 2000 (used on Canada's west coast). Data provided courtesy of Skretting North America<sup>2</sup> Same as g/100 g feed.<sup>3</sup> Protein and fat digestibility are within ranges provided by Skretting. Minerals, phosphorus, fibre and NFE based on Apparent Digestibility Coefficients (Bureau et al. 2003) of salmonids (salmon, charr and trout).<sup>4</sup> The amount of nitrogen is 0.624% (indigestible protein/ 6.25). This is the same as 6.24 g N/ kg feed fed; or 6.86 g N/ kg growth, with a biological FCR of 1.1.<sup>5</sup> Nitrogen Free Extract: primarily carbohydrates.<sup>6</sup> Phosphorus estimates for Optiline 2000 are based on analysis of 11 mm Orion salmon feed (Petersen et al. 2005); a Moore-Clark feed prior to takeover by Skretting North America.<sup>7</sup> This mineral value (ash) does not include phosphorus (row above). The Skretting North America supplied value (all minerals) was adjusted (minus phosphorus) accordingly.

Table 3. Estimates of blue mussel (*Mytilus edulis*) organic faecal loads resulting from the consumption of 100 mt of various dietary sources.

Diet	Dietary OC <sup>1</sup> (fraction)	Dietary Organics (mt)	Absorption Efficiency <sup>1</sup> (fraction)	Digested Organics (mt)	Faecal Inorganics (mt)	Faecal Organics (mt)	Faecal OC (fraction)
Salmon Feed	0.93	93	0.90	84	7	9	0.57
Salmon Faeces	0.77	77	0.86	66	23	11	0.32
Diatom <sup>2</sup>	0.66	66	0.81	53	34	13	0.27
Spat formula <sup>3</sup>	0.77	77	0.87	67	23	10	0.30
TPM <sup>4</sup>	0.36	36	0.54	19	64	17	0.21

<sup>1</sup> Organic content (OC) of diets and absorption efficiencies are taken from Reid et al. (2010).

<sup>2</sup> Commercial diet (Innovative Aqua Products Ltd.) of condensed diatom (*Thalassiosira weissflogii*).

<sup>3</sup> Commercial diet (Innovative Aqua Products Ltd.) composed of concentrated algae (*Phaeodactylum tricornutum*, *Chaetocerus-B*, and *Nanochloropsis oculata*).

<sup>4</sup> Total particulate matter (TPM), randomly sampled adjacent to salmon cage in the Bay of Fundy.

Table 4. Calculations of oxygen demand of IMTA species from a single aquaculture cage. Numbers in parentheses are percentages of the demand from salmon in mussels based on the total oxygen available.

Variable	Value
Ambient DO (mg/L)	8
Cage circumference (m)	100
Cage depth (m)	15
Cage volume (L)	11,936,621
Mean current speed (cm/s)	3
Replacements/hr (based on mean current)	3.4
Total oxygen available (mg/h)	324,000,000
Salmon oxygen demand (mg/h) <sup>1</sup>	12,645,000 (3.9%)
Mussel oxygen demand (mg/h) <sup>2</sup>	19,170 (0.01%)
Total oxygen demand (mg/h)	16,605,000 (3.91%)

<sup>1</sup> Based on the oxygen demand model (Page et al. 2005) where the mass of fish is assumed to be 4.5 kg, the water temperature is 12°C, swimming speed is one body length per second, and the feeding rate is 0.1% body weight.

<sup>2</sup> Oxygen demand was based on a calculated dry weight of mussel biomass of 25 tonnes representing the capacity of one raft (Robinson, unpublished data). Respiration rates were calculated from Vahl (1973).

*Table 5. Calculations of the potential daily organic load of Atlantic salmon faeces during peak biomass in the fall of the year 2 production cycle.*

Description	Value
Number of salmon	300,000
Mean weight at harvest (kg)	4.5
Total weight on farm (kg)	1,350,000
% body weight eaten per day <sup>1</sup>	1.0%
Total food consumed (kg)	13,500
% faeces produced <sup>2</sup>	15%
Daily weight of faeces produced (kg)	2,025
Organic content	77%
Organic deposition (kg/day)	1,559

<sup>1</sup> Consumption rate estimated from Handeland et al. (2008).

<sup>2</sup> Faecal production rates taken from Reid et al. (2007).

*Table 6. Daily projected organic loading rate of IMTA blue mussels grown on 100 m circumference mussel rafts located within an IMTA salmon site.*

Description	Value
Number of rafts	4
Mean weight per raft at harvest (kg)	25,000
Total weight on farm (kg)	100,000
Dry weight of mussels (kg)	13,700
Deposition rate (mg/g dry wt/d) <sup>1</sup>	63.1
Deposition (kg/day)	864.4
Organic content <sup>2</sup>	40%
Organic deposition (kg/day)	345.8

<sup>1</sup> Deposition rates of faecal matter from mussels taken from Callier et al. (2006).

<sup>2</sup> Organic contents of mussel faeces taken from Liutkus et al. (2012).

Table 7. Mussel organic load calculations. OC = organic content, AE = assimilation efficiency, OL = organic load, mt = metric tonnes.

**Example 1: Net reduction in organic load**

Diets consumed	Proportion (%)	Amount (mt)	OC (fraction)	Organic (mt)	AE (fraction)	OL (mt)
Salmon culture solids	17%	6.8	0.93	6.3	0.93	0.4
Natural diet	83%	33.2	0.77	25.6	0.85	3.8

*Model Inputs*

TPM consumed by mussels = 40 mt  
 Salmon culture solids = 200 mt  
 Diet proportion of salmon culture solids = 17%  
 Salmon solids organic content = 0.93  
 Natural diet organic content = 0.77

*IMTA organic load values*

Salmon organic load = 186.0 mt  
 Salmon solids consumed = 6.8 mt  
 Unconsumed salmon solids = 193.2 mt  
 Unconsumed salmon organic load = 179.7 mt  
 Mussel organic load = 4.2 mt  
 IMTA organic load = 183.8 mt

**Conclusion:** Net Organic Reduction = Salmon organic load (186.0 mt) > IMTA organic load (183.8 mt)

**Example 2: Net gain in organic load**

Diets consumed	Proportion (%)	Amount (mt)	OC (fraction)	Organic (mt)	AE (fraction)	OL (mt)
Salmon culture solids	3%	1.8	0.77	1.4	0.85	0.2
Natural diet	97%	58.2	0.4	23.3	0.60	9.4

*Model Inputs*

TPM consumed by mussels = 60 mt  
 Salmon culture solids = 200 mt  
 Diet proportion of salmon culture solids = 3%  
 Salmon solids organic content = 0.77  
 Natural diet organic content = 0.40

*IMTA organic load calculations*

Salmon organic load = 154.0 mt  
 Salmon solids consumed = 1.8 mt  
 Unconsumed salmon solids = 198.2 mt  
 Unconsumed salmon organic load = 152.6 mt  
 Mussel organic load = 9.6 mt  
 IMTA organic load = 162.2 mt

**Conclusion:** Net organic gain = Salmon organic load (154.0 mt) < IMTA organic load (162.2 mt)

*Table 8. Comparison of the perceived costs and benefits of adopting an IMTA approach to a fish farm.*

Potential IMTA Costs	Potential IMTA Benefits
Increased biomass on a lease site, increasing inorganics, oxygen demand and potentially increasing organic loading. There may also be an increased potential for re-suspension and transport of fine particulates away from the site.	Pro-active method for dealing with organic and inorganic nutrient loading to the environment. (Note: that this applies to all forms of IMTA including close containment as well as open water culture).
Increased cost of infrastructure and requirements for continued innovation.	Increases the revenue streams coming from a particular aquaculture site.
Increased system complexity requiring a higher degree of knowledge for monitoring which applies to both operations and management.	Diversifies the products coming from an aquaculture site to reduce economic risk.
Increase in the number and frequency of negative species interactions at the micro to macro levels (e.g. ranging from diseases to predators).	Reduces some of the negative species interactions that are currently occurring that impact fish health.
	Increases employment to service the new additions to the operation at both the company level and the community level.
	Increases the level of understanding of the industry and the environmental managers on ecological interactions that normally take place on an aquaculture site so that they can work more efficiently within certain boundaries determined by the ecosystem. (Note: these interactions were always there, but were not as relevant in the operational decisions in the past).
	Increases the level of engagement with the local communities with regard to infrastructure and public policy.

**Table 9.** Calculations of the mean IMTA site area available on the lease underneath the outside edges of the cage boundaries. Length and width measurements were determined from the compensator floats shown on the aerial photographs of the aquaculture sites using Google Earth™.

Site	Length (m)	Width (m)	Area (m <sup>2</sup> )
Hills Island	122	479	58,438
Man O War	185	476	88,060
Charlie Cove	199	346	68,854
Crow Island	253	366	92,598
Mean area (m <sup>2</sup> )	-	-	76,988

**Table 10.** Projections of biomass and value of potential IMTA species with food calculations based on daily consumption rates of 0.75% body weight per day.

Variable	Sea Urchins	Sea Cucumbers	Scallops
Cage layers	2	5	5
Density (#/m <sup>2</sup> )/layer	300	12	40
Mean weight per organism (g)	50	900	20
Total mass per m <sup>2</sup> (kg)	30	54	4
Area available (m <sup>2</sup> )	2,566	2,566	2,566
Total mass per farm (kg)	76,988	138,578	10,265
Daily consumption (kg)	577	1,039	77
Total number per m <sup>2</sup>	600	60	200
Total number per farm	1,539,750	153,975	513,250

## FIGURES

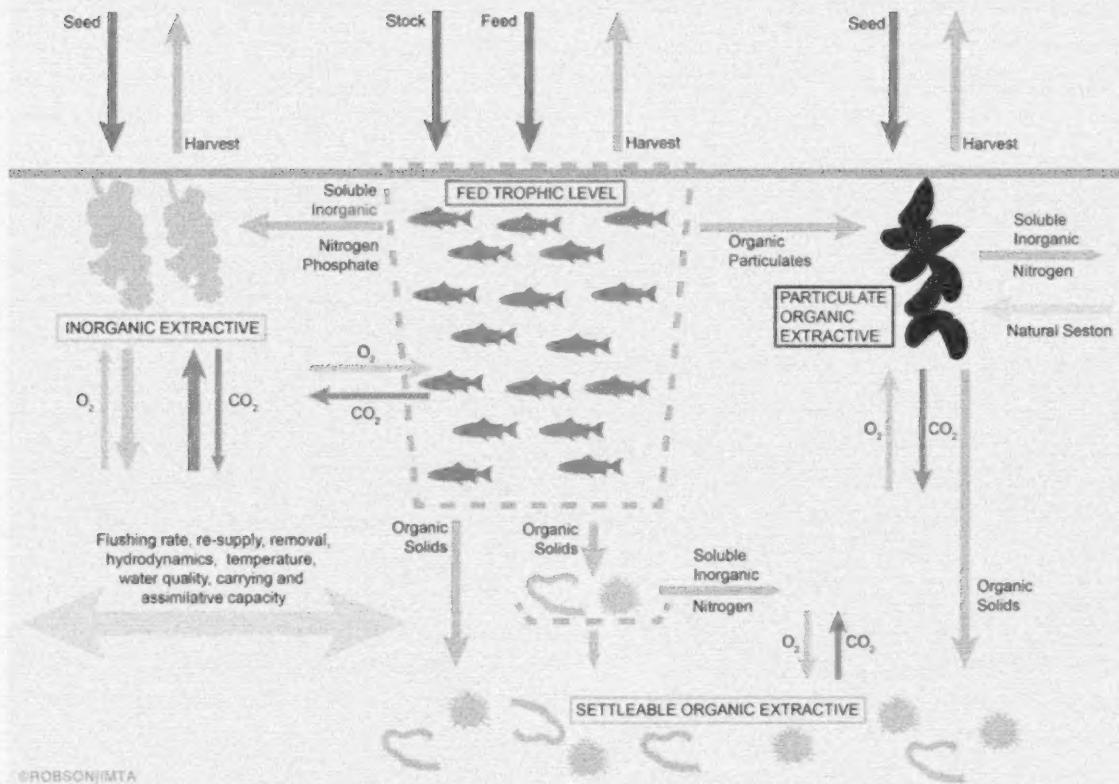


Figure 1. Schematic diagram of the IMTA concept showing the various trophic levels (fed and extractive) and some of the nutrient pathways.

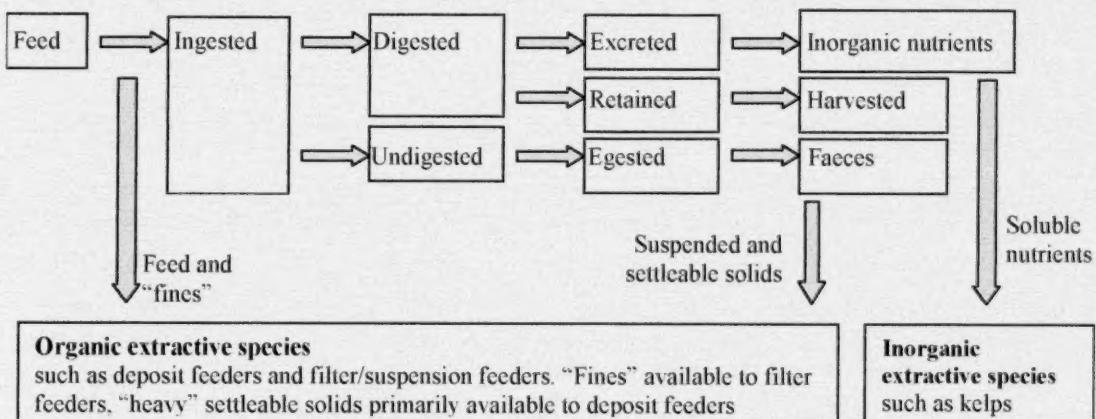


Figure 2. The partitioning of nutrients in salmon production and nutrient losses available to extractive species. Feed that is ingested but not digested will become faeces. Digested feed will be retained in the tissue or metabolized and excreted. Carbon that is digested and not retained is respired as carbon dioxide while nitrogen is excreted primarily as ammonium. Blue arrows indicate nutrient partitioning in fish culture and green arrows indicate losses potentially available to extractive species. Dissolved organic matter (DOM) is also released.

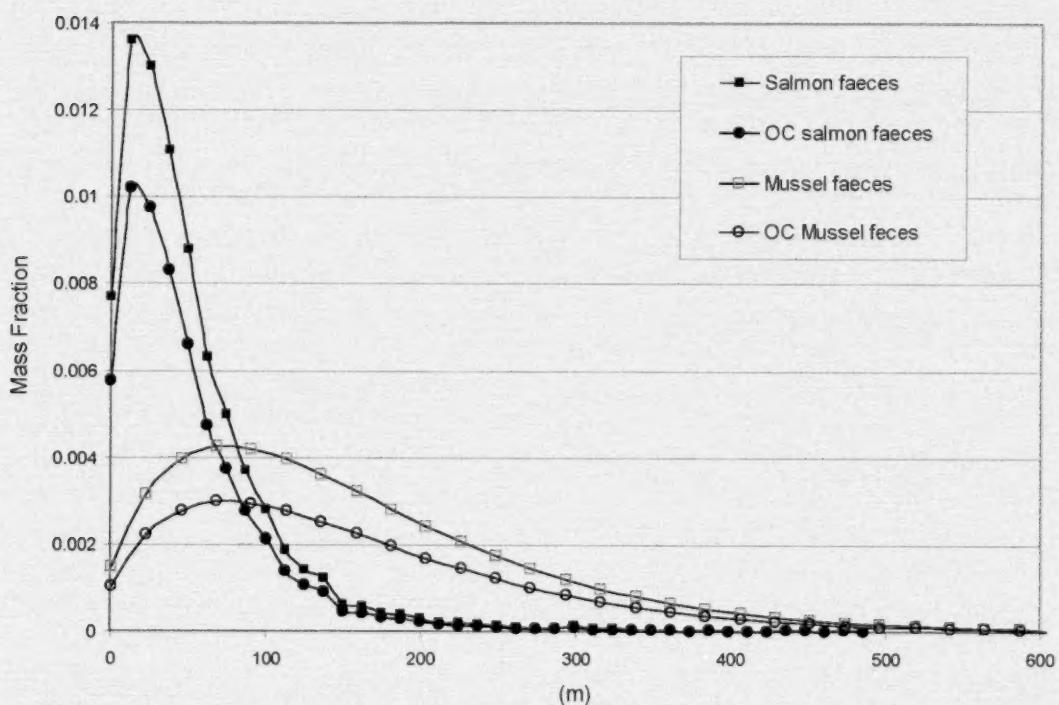


Figure 3. Theoretical deposition of salmon and mussel faeces along a horizontal transect in 20 m water depth. The estimated spread is from an equal mass release of salmon and mussel faeces. Mass fraction refers to the percentage of the organic content (OC) load released. Model development is detailed in (Reid et al. 2013; Appendix 2).

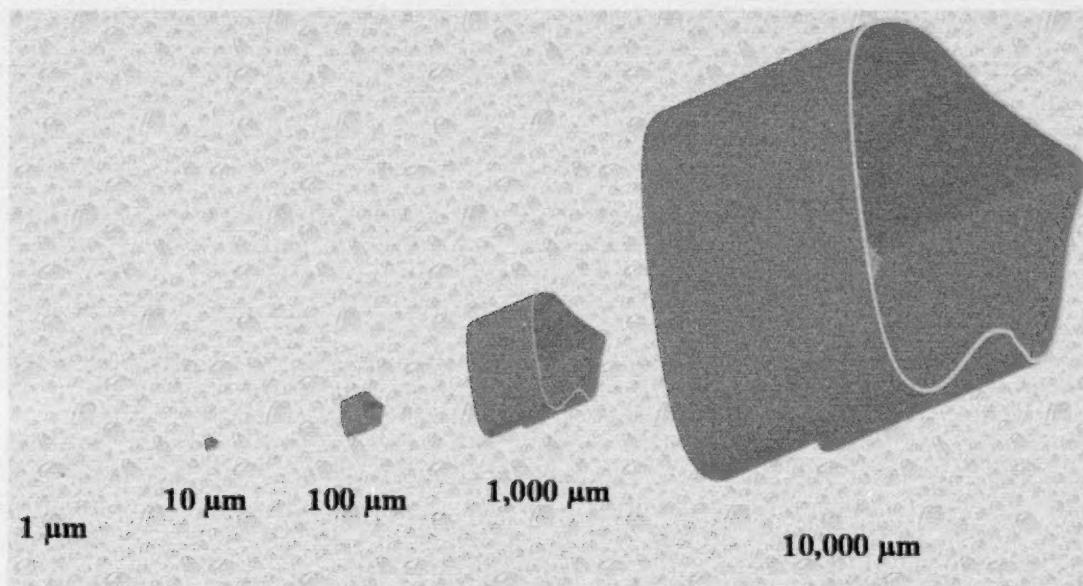


Figure 4. Scale representation of the particle sizes available to filter and deposit feeders growing within an IMTA system. 1  $\mu\text{m}$  particles represent bacteria and small diatoms, 10  $\mu\text{m}$  represents most diatoms and dinoflagellates, 100  $\mu\text{m}$  represents larval zooplankton, 1000  $\mu\text{m}$  represents adult copepods and seston, and 10,000  $\mu\text{m}$  represents salmon food and faecal particles.

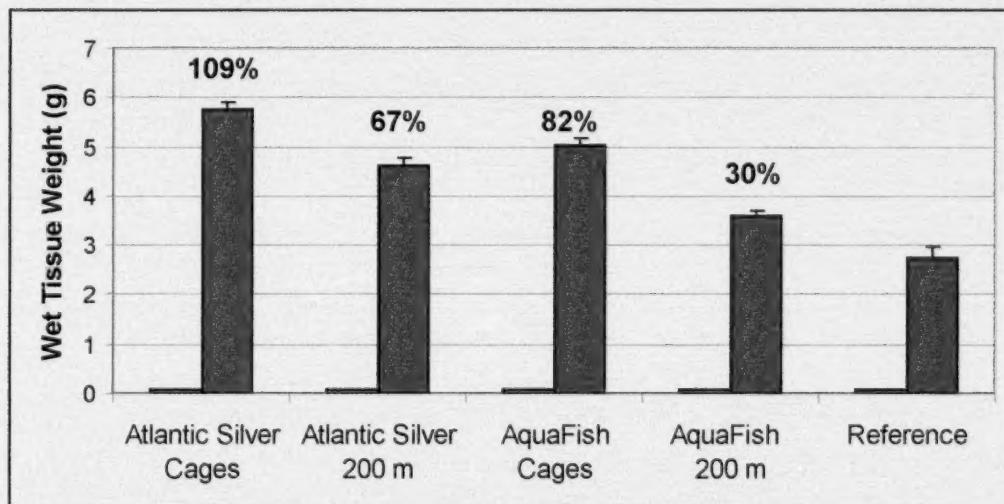


Figure 5. Growth in somatic tissue for blue mussels grown in experimental mussel socks at two salmon IMTA sites in Passamaquoddy Bay from February 2002 to January 2003. The mussels were deployed at the edge of the fish cage or and an independent moorings 200 m distant. An additional reference site was set up 1.2 km away on independent moorings. Numbers above the bars are percentage differences of the weights compared to the furthest reference site.

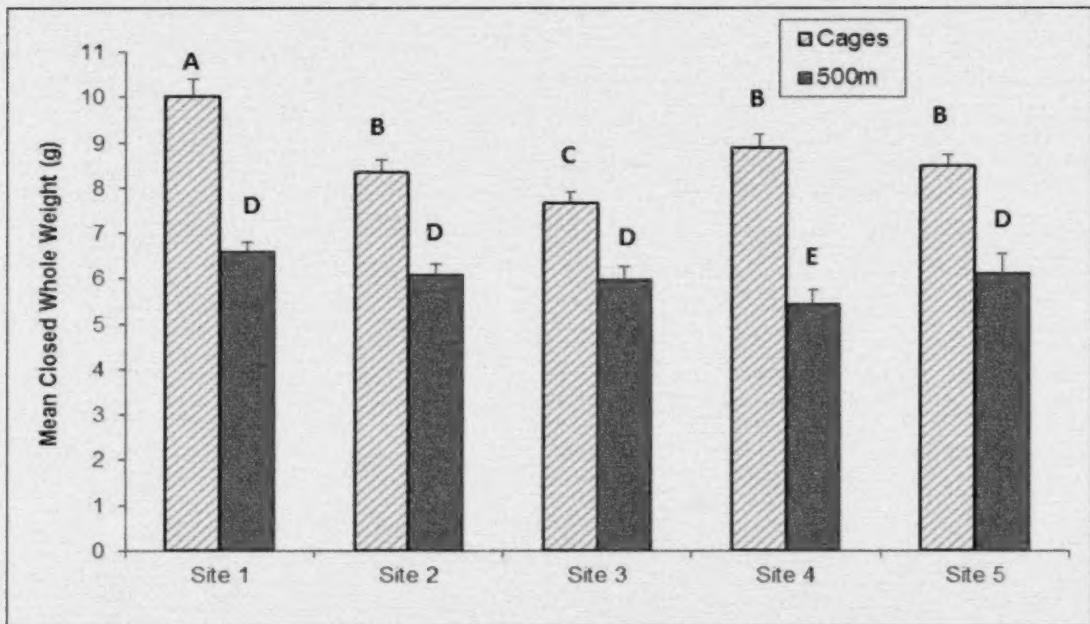


Figure 6. Mean closed whole weight ( $\pm$  SE) for mussels grown at two distances from five salmon aquaculture sites in the southwest Bay of Fundy region. "Cages" = mussels grown directly adjacent to the salmon cages at each site; "500 m" = mussels grown at a distance of 500 m from the nearest salmon cage. Mussels were grown from June to November 2002. AquaFish Farms (Site 1), Aquaventures (Site 2), Atlantic Silver (Site 3), Charlie Cove (Site 4), and J.D. Stewart (Site 5). Letters above the bars indicate the results of a Tukey post-hoc multiple comparison test showing significant differences among locations and sites. (Data from Lander et al. 2012).

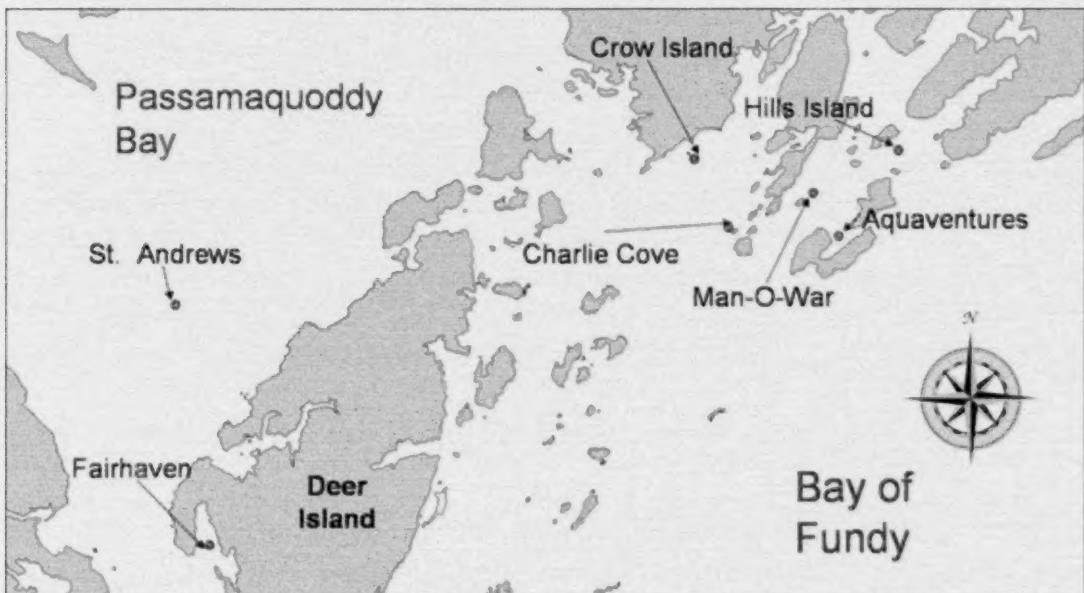


Figure 7. Map showing the experimental IMTA sites used in this study.

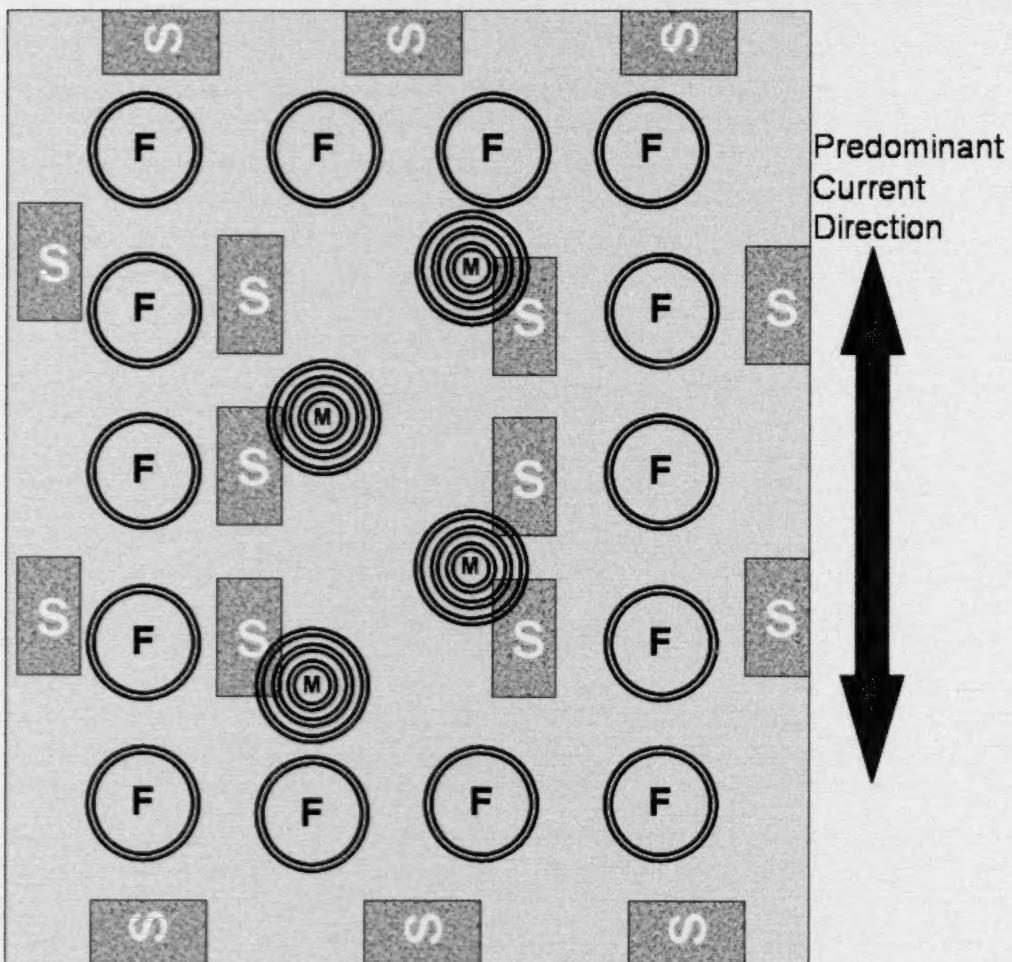


Figure 8. Conceptual diagram of a potential future IMTA site incorporating salmon cages (F), mussel rafts (M) and subsurface cages for other benthic deposit feeders (S).

## APPENDICES

### Appendix 1

#### Estimating the net organic load from an IMTA site with Atlantic salmon and blue mussels

A mathematical model is presented to determine the dietary proportion of salmon culture organics that must be consumed by an IMTA mussel population before there is a net reduction in organic load. A variant of this model is also presented in Reid et al. (2013).

##### Methods

The following mathematical model was developed to determine the dietary proportion of salmon culture solids (faeces and waste feed) that must be consumed by a cultured mussel population to result in a net reduction or net gain in organic load at an IMTA site. There are a number of simplifying model assumptions that should be noted. The model assumes that pseudo-faeces production from the mussels and faecal production from biofouling organisms on the mussels (or associated structures) are negligible. Neither of these scenarios are absent in all cases. Pseudo-faeces can be produced under very high or very low seston concentrations, and heavily biofouled mussel lines can make substantial contributions to benthic deposition (McKinsey et al. 2011). Nevertheless, to assist with model parsimony in the development of first order estimates, these aspects will be excluded in this particular model iteration and this should be considered in the interpretation of model outputs.

A net reduction or gain in organic load at an IMTA site is a function of whether the addition of co-cultured species result in an increase or decrease in organic material relative to what would be loaded as monoculture fish site (Equation 1). The amount of organic material in a particular solid is a function of the organic content of a particular dietary solid (Equation 2). Unconsumed salmon culture solids (and their respective organic content) not filtered by mussels (Equations 3, 4) and are added to the mussel organic load to estimate the total site wide IMTA load (Equation 5). The mussel organic load is the summation of faecal organics produced from all diets (Equation 6). Only two dietary combinations are run in model simulations. The organic load produced from a particular diet is a product of the dietary proportion of all particles (TPM) and the undigested organic content (Equation 7). Absorption efficiency of a particular diet was determined from a regression equation developed from diets of different organic content (Equation 8).

##### Equations

All load calculations are in mass units.

##### Equation 1: Net reduction or gain in organic load

Salmon organic load - IMTA organic load

Where:

$ROL$  = Net reduction in organic load, mass of organic load reduction through IMTA practices

$OL_{salmon}$  = Salmon organic load, mass of salmon culture solids that are organic

$OL_{IMTA}$  = IMTA organic load, organic load from an IMTA site

## Equation 2: Salmon organic load

$$OL_{\text{salmon}} = OC \cdot S_{\text{salmon}}$$

Where:

$OL_{\text{salmon}}$  = Mass of salmon organic load

OC = Organic content, fraction of material that is organic (model input)

$S_{\text{salmon}}$  = Salmon solids, mass of solids (faeces and waste feed) produced from salmon culture (model input)

## Equation 3: Unconsumed salmon solids

$$US_{\text{salmon}} = S_{\text{salmon}} - CS_{\text{salmon}}$$

Where:

$US_{\text{salmon}}$  = Unconsumed salmon solids

$CS_{\text{salmon}}$  = Mass of salmon solids consumed by mussels

## Equation 4: Unconsumed salmon organic load

$$UOL_{\text{salmon}} = OC$$

Where:

$UOL_{\text{salmon}}$  = Mass unconsumed salmon organic load

## Equation 5: IMTA organic load

$$UOL_{\text{salmon}} + OL_{\text{mussel}}$$

Where:

$OL_{\text{mussel}}$  = Mass of organic load from mussel faeces

## Equation 6: Mussel organic load

$$OL_{\text{mussel}} = \sum OL_{\text{Diet}_i}$$

Where:

$OL_{\text{Diet}_i}$  = Mass of organic load produced from consumption of diet  $i$

## Equation 7: Dietary organic load

$$OL_{\text{Diet}_i} = (C_{\text{TPM}} * \text{Diet}_{i\text{frac}}) * OC * (1-AE)$$

Where:

AE = Absorption Efficiency

$C_{\text{TPM}}$  = Mass of total particulate matter consumed by mussels

$\text{Diet}_{i\text{frac}}$  = Fraction of TPM consumed made up of  $Diet_i$

## Equation 8: Absorption Efficiency

$$AE = 0.395 \cdot \ln(OC) + 0.9578 \quad (\text{Reid et al. 2010})$$

Where:

AE = Absorption efficiency, the fraction of organic material digested

A simple spreadsheet model was developed in Microsoft Excel™ with model input requirements of salmon culture solids produced, consumption of TPM by mussels, organic content of salmon solids, organic content of seston, and the proportion of the total diet (TPM) from salmon solids. The model was run testing diets with differing organic content: low quality seston (OC=0.35, measured in the Bay of Fundy), high quality seston (OC=0.77, reflective of pure microalge), salmon faeces (OC=0.77), and salmon feed (OC=0.93). Four dietary combinations of two diets were tested; each with one diet of salmon culture solids and one of seston. Model simulations combined salmon feed with low and high quality seston, and likewise, salmon faeces with low and high quality seston. TPM consumed by an IMTA mussel population and the amount of salmon culture solids were varied in model simulations to determine how these could affect model outputs. However, an upper threshold of one third total dietary proportion of salmon solids was imposed on the model, as it is unlikely that a mussel population will consume more than this, for reasons discussed in this document.

## Appendix 2

### Simulation comparison of mass-fraction settling velocity and organic content of salmon and mussel faeces: Implications for benthic impacts

#### *Introduction*

Different portions of an aquatic faecal load will settle at different rates. This is referred to as mass fraction settling velocities and these can be represented as a distribution. Mussel faeces produced from ingestion of salmon farm organics and natural diets, settle three to ten times slower than salmon faeces and contains approximately half the organic content (Liutkus et al., 2012; Reid et al., 2009). The slower mass fraction settling velocity and organic content of mussel faeces compared to salmon faeces (Liutkus et al. 2012; Reid et al. 2009), suggests an equivalent amount of mussel faeces would theoretically have reduced potential for benthic impact. Certain thresholds of organic carbon deposition from aquaculture are generally defined to be associated with adverse benthic impacts on soft bottoms (Hargrave et al. 2008; Cromey et al. 2002). A slower settling rate should disperse aquaculture faeces farther, reducing the carbon deposition per unit area. In addition, a reduced proportion of organic carbon per unit mass in mussel faeces should further reduce the area benthic loading of organic carbon. Information on mass-fraction settling velocities and organic carbon composition of both faecal types can enable a simulation comparison of benthic carbon loading between salmon and mussel culture. Data from several studies and unpublished field studies are used to model the relative carbon deposition of mussel and salmon faeces along a hypothetical transect, as a means to compare the benthic impact potential between mussel and salmon faecal loads of the same mass, depth and current regimens.

#### *Methods*

The general equation to determine settling distance is detailed below

Equation 1:

$$\text{Settling distance} = \left[ \frac{\text{Depth}}{\text{SV}} \right] \cdot \text{CV}$$

Where:

Depth = Distance to bottom (m)

SV = Settling velocity (cm s<sup>-1</sup>)

CV = Current velocity (cm s<sup>-1</sup>)

Depth was a static value of 20 m a typical depth under fish cages in SWNB. Current velocity, settling velocity of faeces mass fractions from salmon, and mussel faeces were assigned distributions, making the above equation semi-stochastic

#### *Distribution Fitting of Mussel Faecal Settling Velocity*

Mass fraction settling velocities of mussel faeces (Figure A1) were derived from a cumulative mass fraction versus settling velocity (SV) plot of data from mussel faeces collected at two IMTA sites and one monoculture site (with medium sized mussels, 20-55 mm in length) from Liutkus et al. (2009). The theoretical function was fit to the data distribution using Palisade @RISK software. The distribution was assumed to be continuous and treated as probability density data, with SV and mass fraction as x, y coordinates. A least squares approach for fitting was used by @RISK to minimize the root mean error between the density distribution points and the theoretical function (Palisade 2006, Palisade @Risk, advanced risk analysis for spreadsheets. Palisade Corporation, Ithaca, NY). A lower limit fixed bound of 0 cm s<sup>-1</sup> and an upper limit as

bounded but unknown (not infinite), were assumed for the theoretical distribution. Fit quality was ranked by @RISK using residual mean squared error (MSE). The top two ranked fits were Triangular and Beta General distributions, with an MSE of 0.12 and 0.13, respectively. While the Triangular distribution was a slightly better mathematical fit, it was felt that the asymptotic nature of the upper tail in the Beta General distribution was much more representative of natural phenomena than the hard angles of the triangular distribution. Consequently the Beta General distribution was fit ( $\alpha_1 = 1.043$ ,  $\alpha_2 = 1.542$ , min = 0.18, max = 1.95) and utilized for the simulations (Figure A2).

#### *Curve Fitting of Representative Current Velocity*

To obtain a distribution of current velocities (CV) an ADCP (Acoustic Doppler Current Profiler) current record (Paul McCurdy, DFO) from a typical salmon aquaculture site in SWNB was examined. Current records were collected from a depth of 5.5 m every 20 min from June 7 to August 5 (2009) with  $n > 4000$ . CV sample data was fit using @RISK. Sample data was assumed continuous with a lower limit of  $0 \text{ cm s}^{-1}$ . The curve fit was constrained to a lower limit bound at zero and the upper limit as bounded, but unknown. A Beta General distribution ( $\alpha_1 = 1.92$ ,  $\alpha_2 = 32.58$ , min = 0, max = 135.57) was the best fit (Figure A3), of the data distribution (as a histogram) as ranked by Chi-squared, Kolmogorov-Smirnov, and Anderson-Darling fit statistics.

#### *Distribution Fitting of Salmon Faecal Settling Velocity*

Mass fraction settling velocities of salmon faeces was taken from the literature. Stucchi et al. (2005) reported mass fraction settling velocities of salmon faces where the fractions 0.15, 0.70 and 0.15 correspond to 4, 3 and 2  $\text{cm s}^{-1}$  respectively. For the sake of model parsimony the distribution was assumed to be normal. Although this is likely inaccurate, reasonably good predictions of faecal deposition have been achieved assuming a normal settling velocity distribution of salmon faeces (Reid et al. 2009). Consequently, in the absence of more detailed data, a normal distribution was defined in @RISK (mean = 3, variance = 0.693), by setting the means of the lower and upper fractions (i.e. 4 and 2  $\text{cm s}^{-1}$ ) at the .075 delimiter (mid 0.15) of the distribution tails (Figure A4).

#### *Simulation*

A normalized (area = 1) probability distribution was generated to represent the deposition of faecal mass fractions allocation along a hypothetical transect. Settling time was calculated as indicated in equation 1. A depth of 20 m was chosen for simulations. Equations were made stochastic by @RISK using the aforementioned distributions in place of static parameters. Uncertainty in model parameters was represented through the stratified sampling of distributions. In general, the frequency at which a value from an input distribution is sampled and run is proportional to the frequency at which the value occurs in the distribution. This also enables the generation of output distributions (Figure 3). The specific stratified sampling technique used was Latin Hypercube, which accurately recreates the probability distribution specified by the distribution type, in fewer iterations compared to traditional Monte Carlo sampling. Latin Hypercube accomplishes this by assigning the cumulative distribution equal intervals where sampling is forced to represent values in each interval (while randomly sampling *within* the interval) which, in turn, forces a recreation of the probability distribution input (Palisade 2006) with less data demand. Each simulation was run for 10,000 iterations. Filters were applied to exclude deposition outliers further than 600 m, which resulted in excluding 0.3% of the iterations.

#### *Adjustment for Organic Composition*

Relative OC of faeces and sediment was plotted in concert with faecal mass fraction generated by the simulation, to illustrate the proportionate contribution. The theoretical OC of salmon

faeces has been reported as approximately 73% and may vary depending on proximate composition and ingredients in the feed (Reid et al. 2009). In a companion study on the absorption efficiency of blue mussels on diets of salmon faeces, faecal OC from tank reared salmon, (consuming a Shur-gain extruded dry-feed), was measured empirically at 77% (Reid et al. 2010). As it was not possible to collect immediately egested salmon faeces from the water-column at the field sites in this study, a mean value of 75% OC was considered a reasonable compromise for the simulations. Mussel faeces resulting from the consumption of salmon faeces or a phytoplankton based laboratory diet are approximately 30% organic content (Reid et al. 2010) and this value was used to account for the OC from the mussel load. Mean OC of sediment contribution collected in traps at the field sites was calculated by subtracting the mass and organic content contribution from the reference site, for an OC of sediment at salmon farm of 18% and an OC of sediment at mussel farms of 7%. The faecal mass fraction settling distance data was partitioned by @RISK (for plotting in Excel) into 28 and 40 bins for the mussel faecal and salmon faecal data, respectively. The frequency of each bin (equating to the faecal mass fraction at a given distance) was multiplied by the respective OC to plot relative OC of faeces and sediment to illustrate proportionate contribution.

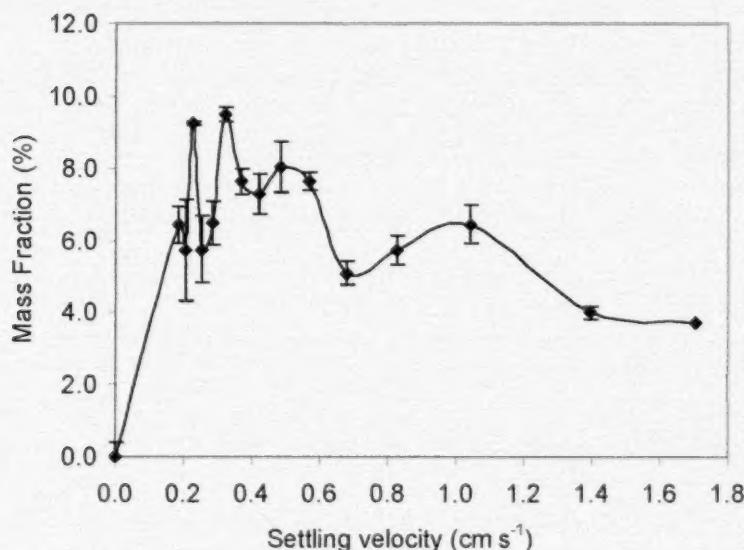


Figure A1: Mass fraction settling velocity of mussel faeces. Each point is a mean of 9 samples. Error bars are standard error.

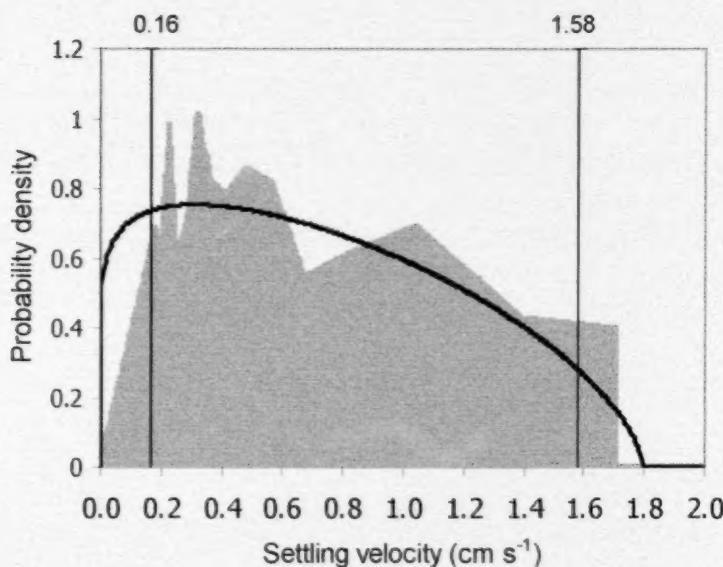


Figure A2: Theoretical distribution fit to mass fraction settling velocity data of mussel faeces. The data distribution is shown in gray and the theoretical distribution fit is indicated by the thick black line. Upper x-axis values indicate the upper and lower 5% data delimiters.

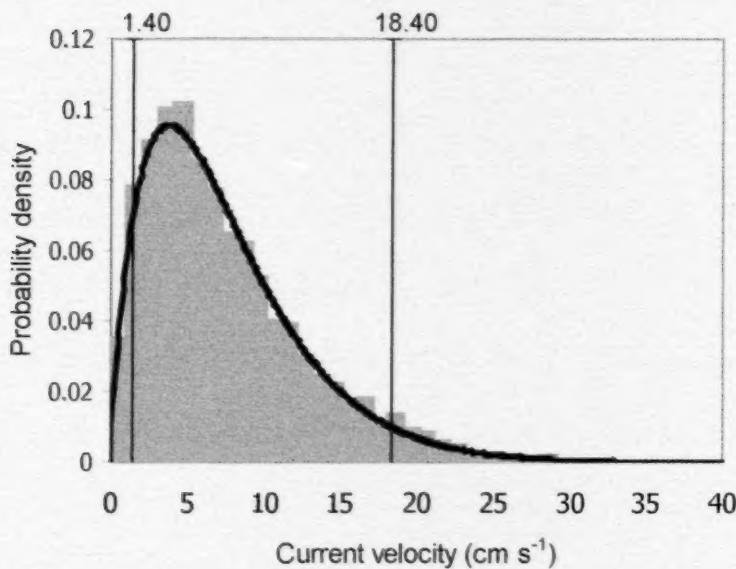


Figure A3: Theoretical distribution fit to current velocity data in Passamaquoddy Bay. The gray areas represent the data distribution while the line is the smoothed fit.

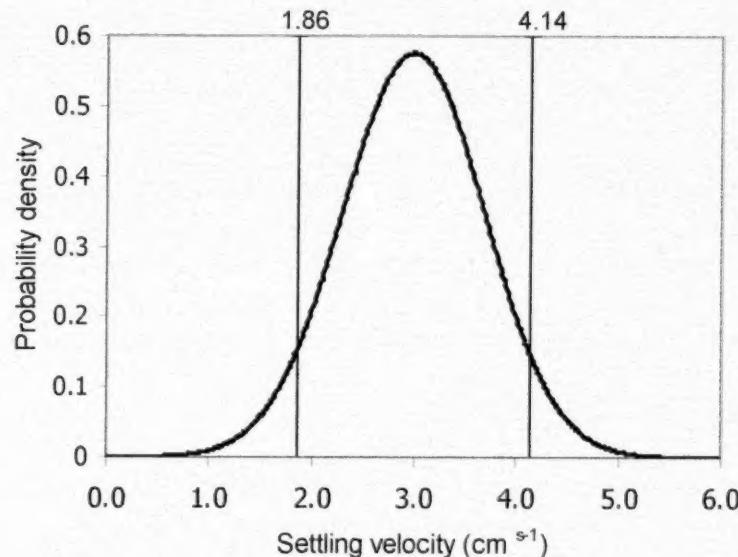


Figure A4: Theoretical distribution of salmon faecal settling velocity. Upper x-axis values indicate the upper and lower 5% data delimiters.